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TEMPERATURE AND LIFE DURATION OF SEEDS

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 226

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(WITH FIVE FIGURES)

Introduction

In this investigation I have sought to determine the extent to which a study of the laws of the life duration of seeds at high temperatures (50–100° C.) will explain the process of degeneration of air-dried seeds at ordinary storage temperatures. In this connection it seemed especially desirable to determine: (*a*) the temperature coefficient (Q_{10}) for the death rate or life duration of seeds; (*b*) to what extent the formula which LEPESCHKIN (**22**) applied as a time-temperature formula for the coagulation of proteins and as a life duration-temperature formula for active living plant cells can be applied as a life duration-temperature formula to dry seeds; and (*c*) how far the temperature coefficient (Q_{10}) on the one hand, and the LEPESCHKIN formula on the other, when applied to actual measurements at high temperatures serve as a means of approximating the life duration of air-dried seeds at ordinary storage temperatures. With these questions in mind a number of determinations have been made on wheat of the Turkey-red variety.

Historical

The effects of high temperatures on dry seeds early engaged the attention of investigators. In 1875 JUST (**19**) showed that dried seeds of *Trifolium pratense* were killed at 120° C., and that

at lower temperatures (time of exposure not given) the speed of germination fell with heating. HÖHNEL (**17**) in 1877 found that most seeds with a moisture content below 3 per cent would endure a temperature of 110–125° C. for 15 minutes. In 1902 DIXON (**13**) summarized the earlier work on high temperatures of seeds as follows: "(*a*) imbibed protoplasm resists 30–40° C. more than the optimum temperature; (*b*) dry protoplasm resists 100° C. more than the optimum temperature of the protoplasm of imbibed seeds." DIXON found that the time required for germination was lengthened as the temperature increased, and that each seed had a heat point at which the time of germination began to increase. In 1899 JODIN (**18**) found that 30–60 per cent of desiccated seeds of peas and cress may be exposed to a temperature of 98° C. without losing their ability to germinate, if first dried at 60° C. for 24 hours and then heated to 98° for 10 hours.

The cause of the loss of viability in old seeds has been a matter of considerable discussion and investigation. DUVEL (**14**) states that seeds retain their viability longest in conditions which permit least respiration, implying that the food materials are exhausted. ACTON (**1**), by a careful analysis of old and new seeds, found that there was but a slight difference in their food content. In the course of these investigations he discovered that there was considerable diastatic and proteolytic enzyme action in new seeds, while in old seeds there was none. He assumed, therefore, that the loss of viability is related to the loss of enzyme activity. The investigations of THOMPSON (**31**), WAUGH (**32**), and others furnish some evidence for this conclusion. They found that old seeds with a low percentage of germination, when soaked in enzyme solution, showed an increase in viability. BROCQ-ROUSSEAU and GAIN (**8, 9, 10, 11**) in their earlier work found that enzymes gradually disappear with age. They tested 300 species of seeds and found no peroxidases in any of the samples secured before the eighteenth century. In the case of *Triticum* they found enzymes in samples as old as 200 years. In some cases the retention of enzymes was attributed to the hard coats of the seeds, and the loss of viability was stated to be due to some cause other than degeneration of enzymes. ASPIT and GAIN (**3**) found enzyme activity in seeds

long dead and in seeds killed by anesthetics. Miss WHITE (33) found no increase in the germination of seeds soaked in enzyme solutions, but rather a decrease due to an increased fungal action. She found the life duration of *Triticum* to be 17 years, with no loss of enzyme activity. According to her work the enzyme theory of the loss of viability is not tenable.

Some very significant work has been done on the time-temperature relation of coagulation of protein both in vitro and in the living cell. BUGLIA (7) found that the time required for the coagulation of blood serum varies with the temperature used. The time of coagulation was found to be a logarithmic function of the temperature. CHICK and MARTIN (12) found that the time required to precipitate egg albumen and haemoglobin from solution varies with the temperature and with the concentration of the solution. LEPESCHKIN (22) showed that the death of active plant cells by supramaximal temperatures is due to the coagulation of the cell protoplasm. He applied a logarithmic formula to express the relation of temperature to the time of coagulation of proteins in vitro as well as in the living cell. By the application of this formula to the determined time for coagulation at any two temperatures, one can calculate the time necessary for coagulation at any other temperature. On this basis LEPESCHKIN calculated the life duration of active *Tradescantia* cells at 20° C. to be 33 days, and at zero to be 3 years. He believes that the life duration of plant cells is very much longer than indicated because of a redispersal process, carried on by the active living cells, which counteracts the coagulation process.

The results of many workers in this field have been well summarized in a recent monograph by KANITZ (20), who has brought together the literature from several related subjects. He shows that in general the effect of temperature upon the rate of chemical processes is governed by the Van't Hoff law, that is, the coefficient for a rise in temperature of 10° C. (Q_{10}) is 2 to 3. From the experimental results at any two temperatures the value of Q_{10} may be calculated from the following equation (referred to as formula 1 and formula 2):

$$Q_{10}=\left(\frac{k_2}{k_1}\right)^{\frac{10}{t_2-t_1}} \text{ or } Q_{10}=10^{\frac{10\,(\log k_2-\log k_1)}{t_2-t_1}}$$

in which k_2 is the rate of reaction obtained at temperature t_2, and k_1 the rate of reaction obtained at temperature t_1.

Many processes in living organisms show a temperature coefficient approximately that of the Van't Hoff law within certain temperature limits. Some of these show high values of Q_{10} at lower or at critical temperatures. High values of Q_{10} are found also for life duration and for coagulation or denaturing of proteins.

KANITZ (**20**) brings out more clearly the relation of temperature to the rate of life processes by recalculating Q_{10} at the various temperature intervals instead of giving only the average coefficient for the whole temperature range. In this way it is found that in many cases Q_{10} is not a constant at all intervals of temperature, but shows decreasing values with rise of temperature.

Method

In order to obtain constant temperatures for heating the seeds, a thermostat was devised as shown in fig. 1. It consisted of an external water bath heated by an electric stove. In this bath was placed a similar vessel of smaller dimensions which was closed at the top and connected with a water-cooled reflux condenser. Methyl or ethyl alcohol or mixtures of methyl or ethyl alcohol with water was used for temperatures 64–99° C. The temperature during the time of an experiment showed a fluctuation of less than ± 0.1 C. For lower temperatures, where the time was much prolonged, the usual water-jacketed incubator was used. This was well wrapped with heavy woolen blankets. The temperature of the incubator was regulated by the automatic electric apparatus devised by LAND (**21**). It gave a very equable temperature, showing a straight line on the drum of an ordinary recording thermometer.

The seeds were heated in the thermostat by inserting securely corked test tubes, each containing 100 selected seeds, through perforations in the top of the inner vessel. These test tubes were suspended by threads passed through the perforations, and the threads were then secured by corks which closed the openings.

Many of these tubes were inserted at the same time and removed in duplicate at successive intervals. Seeds were heated in the

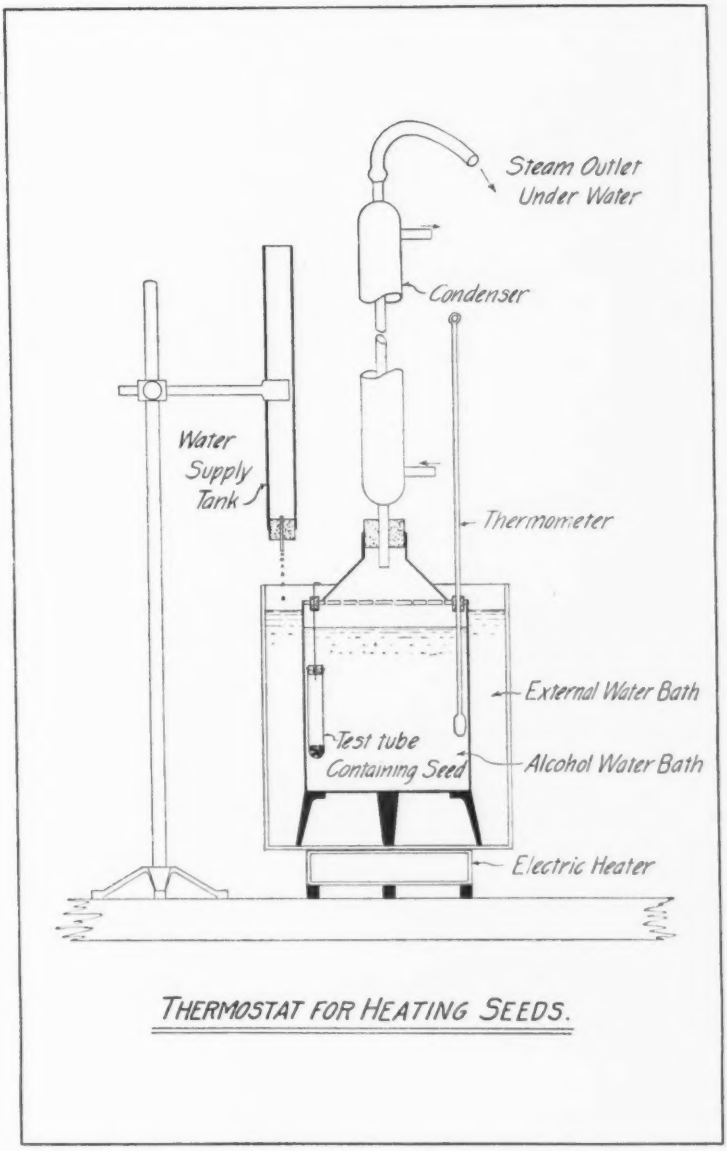
Steam Outlet
Under Water
Condenser
Water
Supply
Tank
Thermometer
External Water Bath
Test tube
Containing Seed
Alcohol Water Bath
Electric Heater
THERMOSTAT FOR HEATING SEEDS.

FIG. 1

incubator in a similar manner by inserting the tubes through perforations in the top. This avoided the main source of temperature fluctuation in the incubator, opening the door.

After the seeds were heated they were sterilized by washing for 2 or 3 minutes in a $n/50$ solution of silver nitrate. While in this solution the seeds were stirred thoroughly in order to free them of all air bubbles. Then they were washed thoroughly in sterile distilled water to remove the excess of silver nitrate which would injure the seedlings when germinated. SCHROEDER (**28, 29**) has shown that the seed coat of wheat is only slightly permeable to silver nitrate, and many parallel tests with treated and untreated seeds confirmed his conclusions. The importance of sterilizing the seeds is realized when we consider that in some cases the germination was delayed as much as 20 days. Miss MÜLLER (**25**), in her work on germination of heated seeds, found that after 10 days seeds were either germinated or destroyed by mold. By sterilizing the seeds and the germinating dishes and using some care in planting, cultures were kept practically free from fungal growth for several weeks.

After sterilizing and washing, the seeds were germinated in large Petri dishes containing a layer of moist cotton covered with a layer of filter paper. The dishes were sterilized at 140° C., and considerable care was used in planting the seeds to maintain sterile conditions. The dishes were kept in laboratory light and temperature, and their daily progress in germination was noted.

The moisture content of the seeds was tested from time to time, and only very slight variations occurred in any one of the 3 moisture content experiments. The DUVEL (**15**) method was used parallel with the ordinary oven drying method and the two gave concordant results. Since the DUVEL method requires less than an hour to make a test, it was possible to check the moisture content before filling the tubes for each trial.

Results

The effects of heating seeds is well shown in table I, which is a daily record of the germination of a time series heated at 87°.5 C. Seeds were considered normally germinated when both root and

stem had broken through the seed coat. When only the root or the shoot appeared, the seeds were considered partially germinated. Partial germination is represented in this table by the figures in small type. The delay in time of germination as the time of exposure increased is strikingly shown here. The controls usually

TABLE I

Record Sheet No. 21 Turkish Red Wheat
Temp: 87.5°C. Moisture 12% April 10, 1914

Time days	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
Control	0	2	2	4	3	3	0	0	0	0	0	0	0	0	0	0	0	0
	92	92	92	93	95	95	98	98	98	98	98	98	98	98	98	98	98	98
7 Min			0	15	12	7	8	7	8	7	5	3	3	4	4	4	3	2
			2	5	27	41	49	55	61	64	67	70	72	72	72	72	73	74
8 Min					4	8	8	7	8	7	5	3	1	5	1	1	1	0
					5	10	25	30	35	41	47	52	54	54	58	59	59	60
9 Min						2	4	4	5	4	5	4	5	2	3	6	5	4
						2	4	8	10	11	18	25	28	32	34	35	37	38
10 Min	Per Cent Partially Germinated									1	3	4	6	4	3	3	4	5
	Per Cent Germinated									0	0	0	0	4	5	9	11	11
11 Min												1	1	0	0	1	2	1
												1	1	2	4	4	4	5
12 Min													2	2	2	2	2	2
													0	0	0	2	2	2
13 Min.																0	0	0

germinate in about 2 days, while some of the treated seeds were delayed for 18–20 days. The relation between time of heating and the percentage of germination is shown in the table. There is a gradual decrease in the percentage of germination with increased time of heating. After the delayed seeds germinate their growth is much slower than that of unheated seeds. It should be noted that the effects here of heating are similar to those produced by the aging of seeds stored at room temperature. This indicates

that there may be a similar change in the two cases. The change occurs rapidly at the high temperature, but slowly at the low temperature.

Some investigators have used the time required to kill all seeds as the end point. In this work we have selected the time required to kill 75 per cent of the seeds as the end point. This is more desirable because there seems to be considerable discrepancy in the resistance of a few stronger seeds. This end point for 12 per cent moisture and various temperatures was obtained as shown in table II. While there are some irregularities, there is a definite relation between temperature and time of exposure necessary for killing 75 per cent of the seeds.

The time-temperature formula suggested by LEPESCHKIN (**22**) has been used here to calculate the life duration of the seeds. By determining the time required to kill seeds at any two definite temperatures, the time for killing seeds at any other temperature can be calculated. The formula (referred to as formula 3) is:

$$T = a - b \log Z$$

in which T is the temperature in degrees Centigrade, Z is the time in minutes, and a and b are constants. If the loss of viability of seeds during storage is a matter of coagulation of cell proteins of the embryo, this time-temperature formula for the coagulation of proteins should be applicable as a temperature-life duration formula for seeds. In experiment the life duration determined must be at relatively high temperatures, ranging from 50 to 100° C. for air-dried seeds.

In formula 3 constants a and b may be calculated by substituting the time and temperature of any two trials and solving for a and b in the two equations. This is the method of calculation used by LEPESCHKIN (**22**). In order to weigh all determinations equally, the constants in this paper are calculated by the method of least squares. The values of the constants and the life duration found in each experiment were substituted in the equation and the theoretical temperatures were calculated. The values are shown in table II. A comparison of these found and calculated temperatures shows that a comparatively close agreement exists. The discrepancies are within the limits of experimental error.

Fig. 2 is a time-temperature curve representing the experimental data shown in table II for wheat with 12 per cent

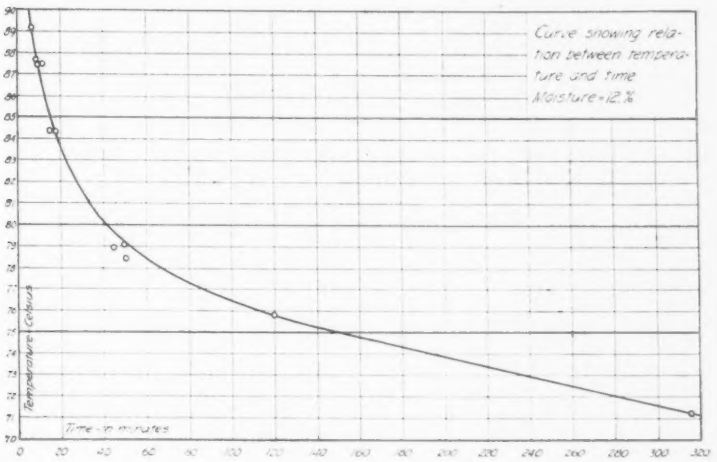

FIG. 2

TABLE II

Germination record of Turkey-red wheat 12 per cent moisture; theoretical temperature calculated by the formula $T=a-b \log Z$; T is temperature (Centigrade); Z is time in minutes; a and b are constants; $a=96.87$; $b=10.4$; value of Q_{10} as calculated by method of least squares = 10.14.

Time in minutes	Calculated temperature	Experimental temperature	Q_{10}
7	88.1° C.	89.2° C.	
8	87.5	87.7	
9	87.0	87.5	6.09
10	86.5	87.5	(10.3)
15	84.6	84.4	
18	83.7	84.4	
45	79.7	78.9	
50	79.2	79.1	
50	79.2	78.5	12.94
120	75.2	75.8	(7.6)
315	70.9	71.3	

Predicted life duration: 50° C., about 22.3 days; 25° C., about 15.5 years; 20° C., about 46.9 years; 0° C., about 393 years.

moisture. The ordinates represent degrees Centigrade and the abscissae minutes of time. Except for the irregularities which

occur at 78 and 79°, there appears a marked agreement in the data and the points representing the experimental data approximate a smooth curve. Table II shows the value of Q_{10} for experimental temperatures as calculated by formula 2. In tables II–IV partial germination is represented by the figures in parentheses.

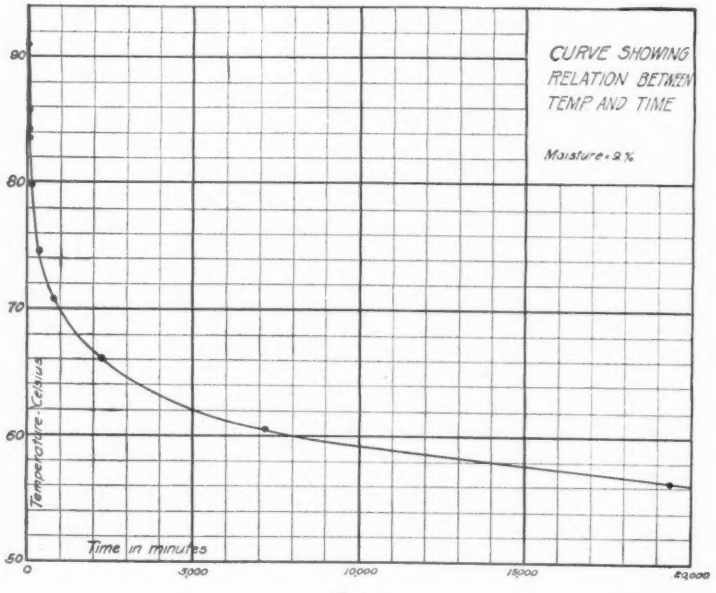

FIG. 3

Table III shows the found and calculated data for wheat with 9 per cent moisture. The calculated temperatures were obtained by the same method as those in table II, and here as before there is a close agreement between the theoretical and observed values. In fig. 3 the experimental data of table III are also expressed in a time-temperature curve. Practically all of the points representing the experimental data fall on a smooth curve. Table III shows the value of Q_{10} for experimental temperatures as calculated by formula 2.

Table IV shows a limited number of data for wheat with 17.5 per cent moisture. As would be expected, the time required to

TABLE III

Germination record of Turkey-red wheat 9 per cent moisture; theoretical temperature calculated by formula $T=a-b \log Z$; T is temperature (Centigrade); Z is time in minutes; a and b are constants; $a=100.8$; $b=10.4$; value of Q_{10} as calculated by method of least squares $=9.23$.

Time in minutes	Calculated temperature	Experimental temperature	Q_{10}
8	91.4° C.	90.8° C.	
27	85.9	85.7	13.49
45	83.6	84.2	(11.0)
63	82.1	83.5	
140	78.5	79.8	
435	73.4	74.4	7.03
810	70.5	70.8	(9.0)
2340(1.6 days)	65.7	66.0	8.51
7200(5.0 days)	60.6	60.6	(10.2)
19440(13.5 days)	56.2	56.3	

Predicted life duration: 50° C., about 53.4 days; 25° C., about 37.3 years; 20° C., about 111.2 years; 0° C., about 938.5 years.

kill such seeds at the temperatures used in the former experiments is exceedingly short. The error due to the time required for the

TABLE IV

Germination record of Turkey-red wheat 17.5 per cent moisture; theoretical temperature calculated by formula $T=a-b \log Z$; T is temperature (Centigrade); Z is time in minutes; a and b are constants; $a=81.73$; $b=8.04$; value of Q_{10} as calculated by method of least squares for last 4 temperatures $=16.45$.

Time in minutes	Calculated temperature	Experimental temperature	Q_{10}
3.0	77.9° C.	87.1° C.	
3.75	77.1	83.6	2.22
4.0	76.9	83.1	(7.6)
5.5	75.8	79.5	
8.0	74.5	74.7	4.90
23.0	70.8	70.5	(9.0)
140.0	64.5	64.4	19.71
440.0	60.5	60.6	(9.9)

Predicted life duration: 50° C., about 6.1 days; 25° C., about 21.6 years; 20° C., about 64.4 years; 0° C., about 2800 years.

seeds to attain the temperature of the bath is therefore very apparent here. The data of this table are expressed also as a time-

temperature curve in fig. 4. Here again there is close agreement between the found and the calculated values. Table IV shows the value of Q_{10} for experimental temperatures as calculated by formula 2.

In fig. 5 the temperature is plotted against the logarithm of the time for wheat with 9, 12, and 17.5 per cent moisture. Since one of the constants is found to have a common value in the 9 and 12

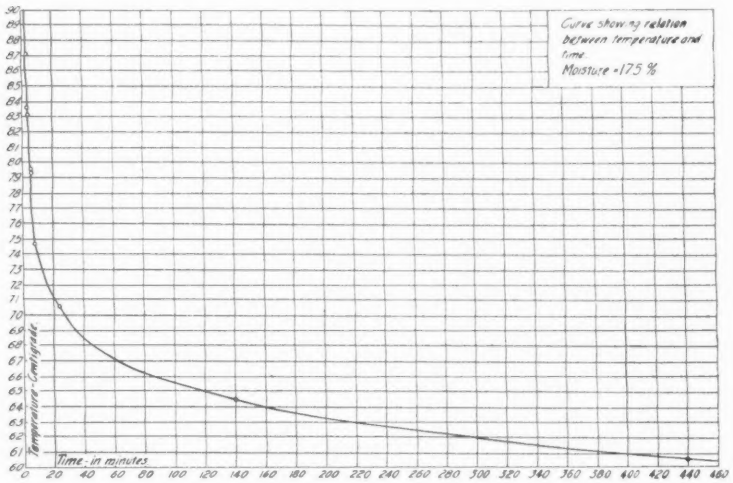
Curve showing relation between temperature and time. Moisture 17.5 %
Temperature - Centigrade
Time - in minutes
0 20 40 60 80 100 120 140 160 180 200 220 240 260 280 300 320 340 360 380 400 420 440 460

FIG. 4

per cent moisture curves, they are parallel. Since they were obtained under identical conditions, one may surmise that the curves in which temperatures in degrees Centigrade are plotted against time in minutes are parallel. Sufficient data are not available with the 17.5 per cent moisture content at experimentally reliable time intervals to justify a conclusive generalization. The curve for the 17.5 per cent moisture content deviates upward from a straight line in the lower time range. This is due to the fact that a considerable part of the short period of exposure was consumed in heating the seed up to the temperature of the bath.

TEMPERATURE COEFFICIENT

The temperature coefficient (Q_{10}) of the life duration of wheat was found to vary with the moisture content. The average value for 9 per cent moisture, calculated by the method of least squares, is 9.23; for 12 per cent moisture, 10.14; and for 17.5 per cent moisture, 9.83. GOODSPEED (**16**), working with barley grains, found a coefficient varying from 10 to 16 as calculated by KANITZ (**20**). The result obtained by GOODSPEED is marred by the lack

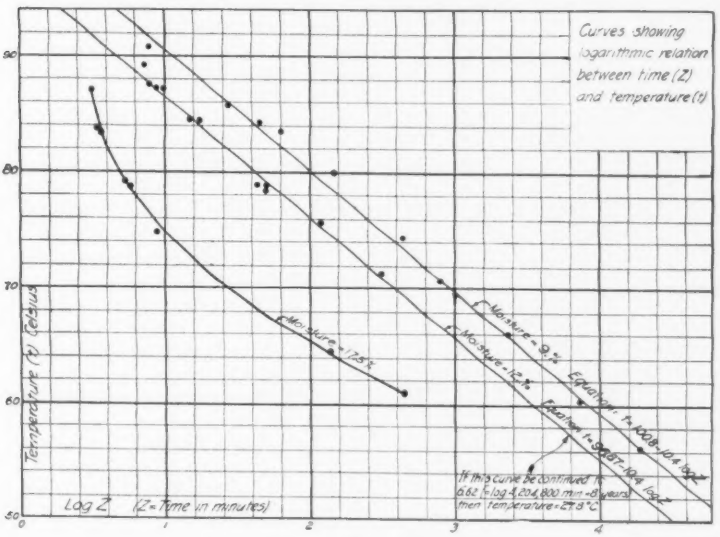
Curves showing logarithmic relation between time (Z) and temperature (t)
90
80
70
60
50
Temperature (t) Celsius
E. Moisture = 17.5%
E. Moisture = 12%
E. Moisture = 9.2%
Equation: t = 100.8 - 10.4 log Z
Equation: t = 99.87 - 10.4 log Z
If this curve be continued it would = 0.82 = log 4,204,800 min = 8 years when temperature = 27.9° C
Log Z (Z = Time in minutes)
0
1
2
3
4

FIG. 5

of determination and control of the moisture content. Since the seeds were soaked for only one hour previous to heating, they had not absorbed the maximum amount of water. Such seeds, when heated in water at various temperatures for various periods of time, show considerable difference in their water content. I find by calculation, according to formula 2, that LEPESCHKIN's data on *Tradescantia discolor* give a value of about 5.0 for Q_{10}, and on *Beta vulgaris* from 2.5 to 10.4. AYRES (**4**), in a recent investigation on one of the red algae (*Ceramium tenuissimum*), found an

average coefficient of 1.47 per degree Centigrade. I have calculated his data according to formula 2 and find the value of Q_{10} to vary from 5 to 88. There is a definite break in his data which prevents them from falling on a smooth curve. This is probably due to some uncontrolled factor in experimentation. I find that the data in other articles cited here, both on life duration and coagulation of proteins, give smooth curves.

The Q_{10} coefficient for life duration of animals, so far as worked, is very much larger than for plants. LOEB (**23**) found a Q_{10} value varying from 240 to 1450 for fertilized sea urchin eggs, while for unfertilized eggs he found a value of 600. MOORE (**24**) found a Q_{10} value varying from 485 to 3900 for the stems of *Tubularia crocea*.

The Q_{10} value for the coagulation of proteins shows a wide variation for the various proteins or different conditions of the same protein as calculated by KANITZ (**20**). CHICK and MARTIN (**12**) found a value of 14 for haemoglobin and 635 for egg albumen. I have calculated the value of Q_{10} for BUGLIA'S (**7**) data and find it to be about 15 for blood serum, 760 for fresh muscle, 45 for neutral albumen, and 240 for neutral concentrated albumen.

The low Q_{10} coefficients found for plants mean that they can endure various supramaximal temperatures for relatively long periods as compared with animals with the larger Q_{10} coefficients. This may be of great importance to plants, since in many habitats they are unable to avoid intense radiant energy. The radiant energy is largely absorbed and the plants attain temperatures as much as 28° C. above the air temperatures, and certainly several degrees above the maximal for growth (**2, 6, 26, 30**). In general the animal is able to avoid such superheating through locomotion. As bearing on this point many more determinations are needed both on animals and on plants of a variety of habits and habitats.

The difference between the temperature coefficients for plants and animals shows itself in an experimental way. There is the possibility of employing a much greater range of temperature in plant material. In the investigations on animals previously cited the range used is about 10° C., while in investigations on plants the range is 20° C. or more. In plants a much greater range

would be possible were it not for the fact that the small coefficients give life durations too long for convenience in experimentation at temperatures not considerably above the maximum.

The difference between plants and animals in the size of the coefficients manifests itself in another way. In the investigations with animals the temperatures used are largely below 45° C., while with plants it is not uncommon to use temperatures above 70° C. and obtain an easily measurable life duration. The maximum temperature usable is determined also in part by other factors, such as percentage of water present and the general attunement of the particular plant to the temperature. The lower the percentage of water in the seed, the higher the temperature that can be used with it. It is probable also that in forms like *Ulothrix* and *Hydrurus*, having maxima below 24° C. (**27**), the possible experimental temperatures would not run so high.

Discussion

The rather close agreement between calculated and found values indicates that the time-temperature formula for the coagulation of protein can be applied as a temperature-life duration formula for seeds, at least under the conditions of these experiments. It is probable that the accumulation of more data will make it possible to find some other equation which expresses more adequately the relationship between the variables involved. In the experiments on wheat of 9 and 12 per cent moisture the average deviation of the observed from the calculated temperatures is less than 1 per cent. The corresponding average deviation for the 17.5 per cent moisture content is about 8 per cent. The unexpectedly large error with the 17.5 per cent moisture content is due to the previously noted fact that a considerable part of the time of exposure is consumed in heating the seeds up to the temperature of the bath. The uniformly increasing deviation of the observed temperatures with short periods of time shows that greater accuracy is possible with long time exposures.

While in many reactions there is a consistent decrease in the value of the coefficient Q_{10} as the temperature increases, we do not find such a trend here. Compared with animal tissue, the value of

the coefficient is small and compares in magnitude with the value found by other workers on plant tissue. The range in the value of the coefficients is small, as indicated by the fact that the data fall on comparatively smooth curves. The coefficient Q_{10} as calculated from the data (for 12 per cent moisture) in table II by the method of least squares is 10.14. When the temperature and time-differences in formula 2 are so small that they are comparable with the errors of observation, then the numerical evaluation of Q_{10} becomes highly inaccurate. But when the time and temperature differences are large enough to render ineffective the errors of observation, then the calculated coefficient Q_{10} is comparable with the value obtained by the method of least squares.

The coefficient Q_{10} for 9 per cent moisture content was found to be 9.23 as calculated by the method of least squares from the data in table III. Similarly, the coefficient for 17.5 per cent moisture content was found to be 16.45 when calculated by the same method, using the 4 highest time observations in table IV. The 4 lowest time observations were ignored on account of the inaccuracy introduced by the time required for the seeds to attain the temperature of the bath, as previously explained.

A number of longevities have been calculated by formula 3 for the low temperatures at different moisture contents. With the relatively short range of temperatures used in these experiments, considerable error may appear in predicted longevities, especially at low temperatures. When such calculated longevities are compared with observed values, they are found usually to be considerably too large, indicating that other processes may also be effective in causing loss of viability. Since hard-coated seeds have long vitality records, it seems quite possible that this is related to the absence of oxygen and low water content.

Much more work is needed to determine how nearly one can thus approximate longevities from measurements made at high temperatures. Determinations should be made on the life duration of seeds with low moisture content. Also similar determinations should be made for a long-lived seed, such as sweet clover, for which we have reliable records of longevity, as well as short-lived

seeds, such as *Drosera*, willow, and poplar. A series of determinations should also be made on seeds at constant temperature with variations in moisture content to ascertain the relations existing between moisture content and life duration.

The data show that the LEPESCHKIN formula applies as a temperature-life duration formula for seeds at the temperatures used in these experiments, but there are several considerations that may limit its application at lower temperatures, including ordinary storage temperatures. (1) Increase of acidity of seeds will hasten the coagulation of the cell proteins; such a change is known to occur in the seeds of certain Rosaceae (**15a**), at least if stored in the imbibed condition. (2) LEPESCHKIN (**22**) found that in active plant cells a redispersal of cell proteins is going on coincidentally with coagulation. As a consequence, at high temperatures where the coagulation was rapid, the found and calculated life durations agree closely; while at lower temperatures, where redispersal is prominent, the calculated life durations are much shorter than the found values. In seeds the calculated values are usually much greater than records of longevity at room temperatures. This indicates that the redispersal process is not going on in relatively dry seeds, or, if it is, it is more than counteracted by some other process. (3) A slight error in the value of the constant b in formula 3 will give a relatively large absolute error for a life duration at low temperatures such as 0° C. At higher temperatures the absolute error becomes less. (4) The lower the water content of seeds, the more heating they withstand and the greater the longevity at moderate and lower temperatures. This law has its limits, for excessive drying is itself injurious. In seeds that will endure desiccation, injury sets in with a reduction of the water content considerably below 2 per cent, while in forms like *Drosera* it appears before air-dry condition is reached. The formula, of course, is limited to degrees of desiccation less marked than those producing injury. (5) It is possible that slow oxidation may limit the longevity of seeds. If this be true, hard seeds with their coats impervious to gases along with their constant low percentage of water are in an especially favorable condition for the

marked longevity which they show. Wheat seeds stored in absence of oxygen might give longevities more comparable with calculated values.

Summary

1. The life durations of wheat with 9 per cent moisture at the various temperatures are:

Life durations in minutes	8	27	45	63	140	435	810	2340	7200	19440
Temperatures in degrees Centigrade	90.8	85.7	84.2	83.5	79.8	74.4	70.8	66.0	60.6	56.3

2. The life durations for 12 per cent moisture are:

Life durations in minutes	7	8	9	10	15	18	45	50	50	120	315
Temperatures in degrees Centigrade	92.2	87.7	87.5	87.5	84.4	84.4	78.9	79.1	78.5	75.8	71.3

3. The life durations for 17.5 per cent moisture are:

Life durations in minutes	3.0	3.75	4.0	5.5	8.0	23.0	140.0	440.0
Temperatures in degrees Centigrade	87.1	83.6	83.1	79.5	74.7	70.5	64.4	60.6

4. The application of the LEPESCHKIN formula at high temperatures as checked by actual measurements gives an average error of 0.6 per cent for 9 per cent of moisture; 0.8 per cent for 12 per cent of moisture; and 8.25 per cent for 17.5 per cent of moisture.

5. The data available for testing the application of the formula at storage temperatures are exceedingly limited. WHITE found that 25 per cent of wheat would grow after being stored for 8.5 years. Assuming that they were exposed to an average temperature of 20° C. and had an average moisture content of 12 per cent according to formula 3, applied to the experimental data of this paper, they should have a life duration of 15.5 years. However, since the variations and averages of temperature and moisture, together with other conditions, are not known, we are not justified in pushing comparisons too far.

6. No definite trend appears in the value of the coefficient Q_{10} and its range is confined to rather narrow limits. For wheat with 9 per cent moisture the range varies from 5.6 to 16.9 with an average of 9.23; for 12 per cent moisture the range varies from 4.8 to 12.6 with an average of 10.14; while for 17.5 per cent the range varies from 2 to 20 with an average of about 10 for the whole scope of the experiment.

7. Since the range of temperature used in these experiments is comparatively short, we are not justified in placing too much emphasis on predicted longevities at low temperatures. Such longevities as have been calculated by formula 3 are large when compared with observed longevities by WHITE and others.

8. This work shows possibilities of throwing some light on the nature of the processes of the loss of viability in seeds in storage conditions. It also makes possible a quantitative statement of the significance of various storage conditions, especially moisture content and temperature, upon the longevity of seeds.

I wish to acknowledge many helpful suggestions by Dr. WILLIAM CROCKER and Dr. SOPHIA ECKERSON, under whose direction this work has been done. I am also indebted to Mr. L. L. THURSTONE of the Carnegie Institute of Technology for aid and advice in making mathematical calculations.

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A REPORT ON SOME ALLOCTHONOUS PEAT DEPOSITS OF FLORIDA[1]

PART II: MORPHOLOGICAL

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(WITH PLATES X AND XI)

Previous to the eighteenth century the question concerning the origin of coal was not debatable, since it was taken for granted that it had arisen as a result of special creation, or during the Noachian deluge by sedimentation (**14**). About 100 years ago, however, the increasing value of coal in the industrial world led many investigators to seek a scientific solution for the problem. One of them, VON BEROLDINGEN, came to the conclusion in 1778 that coal was transformed peat similar to that now found in swamps. After this first step in the right direction, many other students formulated theories as to how the process had taken place. Out of the resulting heterogeneous mass of contentions only two tenets have survived, namely, the allocthonous and autocthonous modes of peat and coal formation. All modern students of the problem are agreed that ancient beds have been produced by an accumulation of organic detritus derived from the old lycopod flora, but there are important differences of opinion still as to the method by which this has been accomplished.

Since detailed and elaborate presentations of the drift and *in situ* hypotheses have found a place in many publications upon the subject, more than a brief review of them would be superfluous in this connection (**8, 9, 10, 11, 14, 15**). Those favoring the first doctrine maintain that these accumulations of much comminuted plant débris, commingled with the more resistant elements, such as spores from vascular cryptograms, carbonized wood (the "mother of coal"), cutinized parts of plants, etc., were deposited very slowly in the bottoms of permanent and open bodies of water, similar to

[1] Contribution from the Laboratories of Plant Morphology of Harvard University.

the method characteristic of lacustrine peat beds. Those opposing this doctrine, the autocthonists, reject the idea that this represents a sedimentation of plant derivatives in open water, but contend that it consists of a gradual amassing of successive generations of lowland plants by prostration *in situ*. The strata, thus exposed, were preserved from decay by a permanent though concealed water supply, as is true and characteristic of the upper stratum of peat deposits in our swamps.

The solution of this vexatious problem, as to which of the two processes is the more probable, has been attempted for the most part by geologists, and naturally enough they have sought explanation topographically. To be sure, the results obtained by numerous investigators in this direction have furnished many valuable data relative to the formation of coal beds, although many of the proofs upon which their conclusions are based are open to serious objection. For example, the presence of stigmarioid roots in coal beds and the supporting shales has been hailed frequently as valuable testimony for an autocthonous origin of the strata in which such structures are found. A broader survey of the problem shows that these rootlike organs are by no means conclusive ground for this deduction, since they also occur quite commonly in cannel coal (a type universally agreed to have been formed in open water), and consequently, a statement which argues equally for either process is unreliable. In this same connection it might be well to mention the quality of the so-called "fire clays" usually found below coal beds. Those who believe that the majority of our coal seams were laid down *in situ* see in the material conclusive proof that this inorganic layer was at one time the subsoil of swamps, owing to an absence of certain minerals which, in their opinion, could have disappeared in no other way than through extraction by growing plants. They fail, moreover, not only to show that this chemical state could not have been brought about by prolonged leaching, but also to account for similar strata in regions which reveal no evidence that they at one time supported forests. In like manner, other topographical features might be shown to present similar objections in favor of sedimentation or an accumulation in place, but this will suffice to illustrate that megascopic investigation alone

is inadequate for reaching substantial conclusions, and therefore it becomes necessary to seek some other means of attack, such as the microscopical study of the material itself. It is evident that the anatomy of coal and peat must be determined before the process whereby they have been formed can be demonstrated clearly.

Studies of the formation of peat, and consequently coal, may be divided into 3 main classes: (1) topographical,[2] (2) ecological, and (3) anatomical.

Topographical features have been considered sufficiently to show that they, especially in relation to coal, are not entirely reliable as a final factor in determining the origin of all classes of organic deposits. The ecology of peat forming plants likewise is limited in its application, and accordingly will receive only a brief consideration, for two reasons. In the first place, literature is quite complete in its descriptions of the usual zones of growth in swamps, including careful enumerations of all species of plants characteristic of them (**2, 6, 10, 11**). In the second place, these plants as such enter but little into the formation of the major part of our peat deposits. Even in swamps only a very small proportion (4–6 per cent) has been derived directly from this flora growing *in situ*. As will be shown later, by far the greater portion of our peat deposits represents a sedimentation of macerated plant material in open bodies of water, and as such is not dependent upon any one zone of growth, but rather upon all indiscriminately. For this reason, the two first named branches of the discussion will not be considered further, except for an occasional reference in connection with the microscopical studies of several characteristic peat beds.

Before discussing any special bog, however, it may be well to introduce a brief description of allocthonous and autocthonous peats as they appear under the microscope. It is possible, of course, to discern with reasonable certainty the methods by which any of our present peat deposits have been accumulated, since one has but to choose his material from clearly defined areas; that is, samples selected from modern lakes present detritus which

[2] FORSAITH, C. C., A report on some allocthonous peat deposits of Florida. Part I: Topographical. BOT. GAZ. **62**:32–52. 1916.

has been deposited in open water, while the very upper stratum of swamps is as equally typical of a cumulative origin in place. A microscopical examination of preparations from open lake deposits shows much minute material, both organic and inorganic. The inorganic constituent may be quite variable, depending upon the character of the surrounding country. If the shores and bottom of the pond are of a sandy nature, and the environment much broken by hills or mountains, a condition favorable for rapidly flowing streams, much sand may be found. On the other hand, if the land is quite level and densely forested, the buoyancy of inflowing streams is much reduced, and the inwash from the shores does not carry any large amount of inorganic material on account of a turflike protection. Consequently, the peat found in such regions will be more or less free from earthy inclusions. This difference in the mineral content of peat in rugged and level tracts is significant, and may throw light upon the topography of coal beds during the period of deposition. All available evidence indicates that the external characters of coal beds were very similar to those just mentioned, inasmuch as the land was flat and heavily forested.

Other inclusions found in peat are the calcareous remains of *Chara*, limy silts, diatomaceous tests, and the shells of mollusks. In addition to these mineral substances, which are small in amount, there occurs the more strictly organic material, derived from more or less macerated portions of plants and minute organisms of sedimentary origin. Some of the most conspicuous of these elements, as well as the most important from the scientific standpoint, are pollen grains of the Abietineae and catkin-bearing angiosperms, and spores from ferns, fungi, etc., representing bodies quite analogous to the microspores and megaspores so habitually found in coal. It is especially important to note that normally autocthonous peats do not show the characteristic spore content so universally found in open water formations. In addition to this microspore material, one finds upon an examination of lacustrine samples a rather large volume of amorphous material. Imbedded in the flocculent matter, there appear ingredients the form of which is more intact, such as woody and herbaceous plant fragments, idioblasts from water lily stems, strips of cutinized epidermis, etc.

Besides these plant remains there are certain animal derivatives characteristic of allocthonous peat; for example, ejecta from fish and small aquatic animals, often containing pollen, diatoms, and bacteria; chitinized portions of insects; spicules from fresh water sponges; infusorial bodies; and shells of mollusks and protozoans.

In contrast to the usual inclusions in allocthonous peat (pollen, diatoms, spicules, idioblasts, etc.) there is the strictly autocthonous peat composed of more or less disorganized plant débris. A superficial examination of this material shows a light brown fibrous or dark brown granular texture, depending upon whether or not the included plants are herbaceous or woody. If the substance is more completely decayed, owing to prolonged maceration and the action of fungal enzymes (unhindered by a constant water covering as is true of allocthonous layers), the fallen plants may become so structureless that they resemble humus rather than peat. Under the microscope this form appears quite homogeneous in contrast to the more fibrous and less decayed *in situ* peats, but seldom do the distinguishing features of lacustrine peat appear, only a tangled mass of roots, stems, and leaves in all stages of decay.

Thus it is apparent that there are two distinct types of peat, presenting structures each peculiar to itself, dependent upon the mode of deposition. If a specimen of coal, therefore, can be shown to present a structure analogous to either of these two more recent formations, it is but natural to assume that its composition is due to similar processes of deposition. Strangely enough, this has not been the usual mode of reasoning. Although it is authoritatively asserted that by far the greater number of the peat deposits in the United States are allocthonous in origin, a diametrically opposite view is maintained in respect to the genesis of coal beds. Consequently it will be the object of this paper to show (1) that these two types of peat are microscopically distinct; (2) that some of the bogs (especially swamps) are not, as is usually believed, of *in situ* derivation, but filled lakes in which the peat mainly represents the lacustrine or open water phase; and (3) that coals in general show clearly the organization of allocthonous peat.

The methods used in carrying out these investigations were as follows: Samples of different types of peat were carefully chosen

from localities throughout a wide range extending from eastern Canada to Florida. The numerous deposits were so selected that all stages in the formation of peat beds were inspected, including large and small deep lakes with sandy shores; filled lakes where the zones of growth have entirely covered the former body of water with a layer of accumulated vegetation; large and small shallow lakes; and swamps and river estuaries. At every station a vertical series of samples was secured at 1 ft. intervals. The probings were made over a sufficient area to allow an estimation of the depth and extent of each deposit. The specimens were obtained by the use of a probe devised by DAVIS (3), and stored in cloth sacks. A careful record was made concerning the topography of the region, the gross character of the material, and the depth from which it was taken. These specimens were later studied microscopically in order to determine the correlation between the grosser structures and the minute anatomy in respect to the mode of deposition.

Turning to the more detailed consideration of the several progressive steps in bog formation, Lake Weir in Florida may be considered as an example of the first stage. Sandy shores surround this body of water, and probings show that there are no accumulations of organic detritus nearer than 100 yards off shore; while beyond this there appears a quite extensive stratum of lacustrine peat. A gross examination of the material showed a consistent homogeneous mass, the grayish color of which is due to a calcareous silt. In addition, there appears a very large amount of diatomaceous and limy remains of extinct plants and water animals. The more peatlike content manifests itself as pollen of abietineous and amentiferous derivation, amorphous matter, root fragments (the stigmarioid rootlets so characteristic of certain samples of coal), and herbaceous and ligneous elements from the higher plants.

Attention may now be directed to the more organic peats in order to establish their relation to coal more definitely. In the first place, I shall consider two forms, the one modern and the other ancient, the origin of which is undoubted, namely, lake peat and cannel coal. Samples of lacustrine "muck" were found in the centers of Lakes Newman, Orange, Griffin, Harris, Apopka, Eustis, and many others in Florida, New Hampshire, Massa-

chusetts, and eastern Canada. In general, the samples obtained by probings were deep brown and plastic. They were fine and uniform in texture, without large or fibrous inclusions. As the topographical features around Lake Harris, in Florida, dispel any doubt as to the allocthonous genesis of the stratum there found, preparations from it will be discussed in detail. Fig. 1 represents a sample taken from near the top, and a careful study of it shows clearly pollen grains imbedded in an amorphous mass of drifted and windblown floatsam, ejecta from water animals, etc. A small spore may also be observed. Sponge spicules and idioblasts from water lily stems likewise appear.

A deposit very similar to the one just described was found in Lake Dot, a small dumb-bell shaped body of water near Eustis. This lake is very interesting as an example of those deep bowl-like depressions, known as "lime sinks" (**12**), which are caused by a subterranean solution of the underlying limestone, so that the roof, becoming too thin to support its own weight, falls. Fig. 2 illustrates a section 3 feet from the top of a 9 ft. layer, and the characters pictured were found by a study of the entire series to be uniform throughout. It will be seen that this sample presents the usual structureless material, ejecta, idioblasts, and pollen. In fact, such structures as are usually encountered in lake "mud," but absent from autocthonous deposits.

The central layers of the peat in Lake Eustis furnished the material shown in fig. 3, which shows several diatoms of the *Stauronesis* and *Navicula* type, in addition to spicules from decayed fresh water sponges. The section also shows 3 specimens of the amoeboid *Arcella*. Other features already found to be characteristic of lake precipitations are idioblasts, pollen, etc. Fig. 4 represents a sample of peat much like those just considered, except that there are more plant fragments. The section from which this sample was taken depicts the type of peat found in Lake Orange a mile off shore, and the topography of the region, as well as the microscopical structure of the material, shows it to be of undoubted lacustrine origin. Although many other deposits throughout a wide range were studied, these 4 illustrations are sufficiently characteristic of all deep water formations, as well as the lower layers of bogs, to

demonstrate the distinctive features of all such lacustrine accumulations.

It is at least significant that cannel coal, which is universally considered to be of open water derivation, should manifest the same structures so generally found in lake peat. Both of these fuels, when microscopically examined, present a considerable spore content. In fact some of the cannels (especially tasmanite) as well as their modern homologues were found by JEFFREY (7) as a result of studies of a great number of carefully prepared sections to be almost entirely sporiferous. A somewhat clearer idea of this correlation may be obtained by a reference to fig. 5, which exhibits the organization of Kentucky cannel coal as it appears under the microscope. Scattered throughout the section there may be seen numerous light bodies, which are the flattened spores of vascular cryptogams (homologues of the spores and pollen shown in the illustrations of allocthonous peat). The long grayish bands are indicative of metamorphosed bits of wood. Separating these spores and lignitoid fragments are dense black masses of amorphous organic material. In comparing this illustration with fig. 4, it is apparent that both the recent and prehistoric deposits show a striking anatomical similarity. Thus it would appear that whenever a peat and coal show like organization it has been brought about by the same methods of deposition. It will consequently be assumed in the sequel that similarity of structure, as between peat and coal, implies an identical mode of origin.

Since the next stage in peat formation is illustrated by those areas where the zones of growth are starting to form around the shores, an example will be given. The shores around Lake Orange in Florida show this fringe of water plants quite well. The peat derived from this vegetation consists of fragments of amphibious angiosperms, among which rushes, water lilies, pondweeds, etc., are common. Although the plants which are found in this zone vary systematically in wide ranges, the peat formed by them is uniform. Since the parts of plants which enter into the composition of this peat are very minute, specific differences are not of importance, and consequently any enumeration of them is omitted. Although it is allocthonous, samples taken from this deposit are of

a light brown spongy nature. Fig. 6 presents a preparation from this material, and it will be seen that there are many parts of plants in a perfect state of preservation due to a perpetual covering by water. Other structures more definitely related to lake peats are idioblasts and parts of insects.

Florida is especially favorable for studies of this type of peat, owing to an abundance of "saw grass" (*Cladium*) marshes about Lakes Harris, Griffin, Apopka, and in fact generally throughout the Everglades. If one were to rely solely upon a superficial examination of this material, representative of the later stage in herbaceous marsh development, he would reach the conclusion that these deposits have been formed by a growth of herbaceous plants *in situ*. A detailed examination of samples from different depths, however, shows that this is not a correct interpretation. On the contrary, these paludal accumulations, with the exception of the uppermost layers, are obviously allocthonous. A sample secured 3 ft. from the bottom of the marsh bordering Lake Harris is pictured in fig. 7. This illustrates conclusively that the material has not been formed *in situ* by a gradual amassing of fallen plants, but rather by a floating together of drifted and wind-blown matter similar to that characteristic of deeper lake deposits, as indicated in figs. 1–4. The usual structures found in lacustrine peat, shown in fig. 7, are pollen grains, idioblasts, plant fragments, ejecta, and formless drift. Although no sponge spicules and diatoms appear in the illustration, it should be added that they are of common occurrence. This kind of peat is usually encountered in the lower four-fifths of "saw grass" marshes, as determined by vertical series of samples. The upper layer, nevertheless, has been accumulated in a different manner, since the microscope reveals only the tangled remains of fallen herbaceous plants, and the structures usually found in open water deposits are conspicuously absent. It is probable that this distinct change in the process of deposition was accomplished at some time when the material had so collected that the mass was above water, for a part of the year at least, so that plants perishing in place were allowed to become more or less reduced owing to exposure, and not permitted to float away and become precipitated among the usual sedimentary detritus. It is

apparent that this peat, a very common type in the Florida lake region, is not, as is ordinarily supposed, of autocthonous derivation; but, on the contrary, is almost entirely allocthonous. It would thus appear that the development of this form of deposit is in accord with the general principle of sedimentation for peat and coal in general.

One of the most interesting phenomena in relation to the formation of peat beds is that illustrated by completely or nearly filled lakes. As has previously been stated, there are several distinct steps in the process, beginning with an open lake surrounded by sandy shores, of which condition Lake Weir served as an example. The next is seen where the herbaceous zone has crept in from the shores, as illustrated by the "saw grass" marshes around Lake Harris. The third stage is the conversion into a bog as a result of drainage and the introduction of woody plants, which marks the end of the process. Consequently, the value of this last formation as a peat builder has in all probability been overestimated, since the detritus formed by it directly comprises but a small proportion of the whole, especially in the more tropical areas where perpetual exposure is favorable to destructive activities. A series of samples from one of these beds, if studied only superficially, shows two distinct types of material: the upper layer consisting of a tangled mat of fallen plants and roots, and the lower layer consisting of a somewhat homogeneous mass of minute débris. This older plastic material is believed by many writers to have resulted from a more prolonged period of reduction of detritus similar to that found in the upper part of the bed. This conclusion, derived from gross examinations alone, is nevertheless misleading, and on this account it seems advisable to refer to microscopic investigations. A bog near Leesburg will serve to illustrate. A topographical study of this area showed that it was at one time either an arm of Lake Harris or a connecting link between Lake Harris and Lake Griffin. At the present time the filling processes have reached completion, and the entire area is now dry land bearing a dense forest of coniferous and deciduous trees. Probings in several localities showed about 15 ft. of peat resting upon a stratum of bluish clay (the initial stage of "fire clays" usually found under coal beds). Above this lamina

there occurs a layer of fine black peat, similar in form to that now found in the open lakes. This is the "completely decomposed stratum" just mentioned. Fig. 8 shows a microscopical section taken 3 ft. from the bottom, and further studies of the figure reveal in addition to the usual structureless drift, woody and herbaceous plant fragments, pollen, spores, spicules, etc., all of which have been preserved from decay by a perpetual water cover and natural acidity. It is manifest that this material does not represent the final stage in the reduction of fibrous peat, but rather an accumulation of drifted and wind-blown matter which was precipitated at some time when lacustrine conditions prevailed. This relation is still more obvious when it is demonstrated to be similar in the most exacting detail to that already shown to be characteristic of present lake deposits and illustrated in figs. 1–4. Above this dark amorphous mass, there appears a light brown fibrous material like that already described for the "saw grass" marshes. Fig. 9 shows photomicrographically the true nature of the substance. In addition to root fragments across the illustration, there appears the usual disorganized material, pollen, spores, and spicules, all of which indicate an allocthonous origin. An even clearer idea of the lacustrine nature of this peat may be obtained by reference to fig. 10. In the upper right hand corner of the figure is a much distorted fragment from some herbaceous plant, amorphous matter, and ejecta. The most noticeable, as well as one of the most significant, features, however, is a fern sporangium and a sponge spicule which could not occur in juxtaposition except through sedimentation in open water.

Microscopical studies of this vertical series indicate that about the time when the last of the herbaceous material had been deposited, the accumulated mass was above water level, thus furnishing a somewhat drained soil for the growth of more woody plants. Consequently the amphibious species were forced to move on, and their place was taken by woody trees and shrubs. This later growth in turn built up a layer of autocthonous peat which shows the remains of comminuted material, but none of the structures so characteristic of the allocthonous layers below. In securing these samples some difficulty was experienced in forcing the probing

instrument through the tangled cypress logs and roots, which resisted decay more than the dicotyledonous trunks and settled through the oozelike mass below. Although these structures are not general in peat beds, they are by no means uncommon, and in all probability have homologues in coal beds, a fact which has led to the idea that they are indicative of an autocthonous origin for coal. Like all other megascopic evidence, however, the interpretation of these structures is open to question, since conditions like those in the bog just mentioned might have prevailed in the past, and fallen logs settled through the unresisting lake peats below the growing stratum.

It must be apparent that allocthonous peats in this region are vastly predominant over those laid down in place, which is quite in accord with the statement of DAVIS (**15**), namely, "the fact [is] that at the present time peat deposits of this type [lacustrine] are numerically more important than any other in regions where peat formation is common." The even more pronounced dearth of accumulated generations of plants *in situ* in this region than is usual in the more northern bogs is without doubt due to climatic conditions, which in warmer localities are more favorable to the destructive action of fungi. Since Florida now has a climate similar to that generally ascribed to the coal-forming periods, it seems logical to infer that strictly *in situ* depositions were equally scarce during ancient times. This phenomenon is well illustrated, in fact, by several swamps in Florida where the sandy floors do not present any quantity of autocthonous peat. For example, in an extensive swamp near Gainesville there appears a dense growth of cypress, pine, and dicotyledonous trees growing up through an almost impenetrable tangle of fallen trunks in all stages of decay. One can hardly imagine a more favorable location for the accumulation of autocthonous peat, but in spite of this, an examination showed but a few inches of humus-like substance derived from comminuted plants.

Although these Florida peats present conditions of environment more like those which formerly prevailed over the entire earth, some attention should be paid to the more northern deposits, as in all discussions of the problem of coal formation, they are mentioned

as the counterpart of "autocthonous" coals. A bog of this type near Fresh Pond in Cambridge, Massachusetts, will serve as an example. Samples were taken in the usual way throughout the entire 30 ft. of the deposit. With the exception of the extreme upper stratum, the samples present a uniformly brown plastic consistency, similar to that found in open lakes. A subsequent examination revealed that the lower 28 ft. were singularly constant in respect to structure, and composed of the usual amorphous material in which were imbedded pine, larch, and amentiferous pollen; spores and sporangia of ferns and fungi; vast quantities of diatomaceous tests and sponge spicules; minute fragments of roots, stems, and leaves of the higher plants; and animal derivatives such as insect parts, water organisms, and ejecta from aquatic creatures. All of these structures are very similar to those shown in figs. 1–4, with the exception of unimportant northern and southern floral differences. There also appeared in this layer some indications of carbonized woody fragments which had been washed into this former lake from a region swept by a prehistoric forest fire, and there deposited. This fact is significant, since such structures are of common occurrence in coal sections in juxtaposition with unburned material, precluding the possibility of deposition *in situ* (7). These inclusions, together with the wonderfully perfect preservation of the débris even in the very lowest strata, dispel any doubt that it is of an allocthonous origin, and not one brought about by an accumulation of fallen plants which later decay to a structureless mass (the "completely decomposed peat" of many writers).

The next swamp to be considered is a so-called "*Sphagnum* bog" in Auburn, New Hampshire. This deposit is found in depressions between long irregular ridges, in the form of the letter Y, about 2 miles in length and half a mile in breadth. In the central portion there occurs a chain of more or less circular ponds surrounded by the usual zones of growth, the inner zone of which is distinctly sphagnoid. A series of tests showed a layer of lacustrine peat about 27 ft. in depth. Above this and near the ponds there is a floating "mat" of *Sphagnum* and other plants about 1 ft. in thickness. Back from the shores there appears a tangled mass of roots and fallen plants above this mossy stratum. Excepting

this thin autocthonous deposit, all the microscopical sections showed "muck" formed by sedimentation similar in composition to that already described for the bog in Cambridge and in more southern regions. Material from several lakes in eastern Canada was minutely examined, and structural evidences of allocthonous peat were found to correspond so closely to those in the United States that any further discussion of them is unnecessary.

Since the shallow or intermittent lakes are not favorable to an accumulation of any appreciable amount of peat, owing to periods of drought and constant agitation by waves, they will receive but brief consideration. Many of this type were observed around Zellwood and Lake Tohopikaliga in Florida, and several small ponds in New Hampshire. In all there was little peat, especially in the south, where there are distinct wet and dry seasons favoring the destruction of whatever material may have gathered during periods of inundation.

There now remains only one distinct kind of fresh water peat to be considered, namely, that found in river estuaries. One example at Pablo Creek near Jacksonville, Florida, will be discussed. Topography indicates that the space between two elevations was once occupied by a river a mile in breadth. This broad stream gradually filled its bed with organic material until the mass had sufficiently accumulated so that the entire depression was covered by an allocthonous layer of peat, excepting the channel of a meandering river. Tests showed a uniform deposit about 12 ft. in depth. Fig. 11 shows the microscopical character of the material, and studies of the entire vertical series indicate a general uniformity. It will be observed that coniferous woody fragments are very abundant, as indicated by a uniseriate ray in tangential section and an absence of vessels. There are also present the more evident lacustrine derivatives, such as a broken sponge spicule, pollen, amorphous material, and a group of spores. These structures indicate that the deposit, like those found in the now open and filled lakes, has arisen by similar processes of deposition in open water.

The peat illustrated in fig. 11 is especially favorable for comparison with thin sections of the more lignitoid coals (bituminous)

as shown in fig. 12. This pictures a microscopical section of bituminous coal from Perry County, Ohio. Crinkled bands of compressed wood are especially distinct in the upper and left hand parts of the figure, and evidences of an allocthonous origin for this coal are present as flattened spores, appearing as light bodies imbedded in a dense black amorphous matrix. Both the lignitic coal and the woody peat show a large and varied amount of xyloid material in addition to the more obviously lacustrine derivatives, such as spores, etc., which have been shown to be characteristically absent in "swamp" peats. It seems logical to conclude, therefore, that these substances so alike in structure must have arisen by a similar process, and for this reason any coal showing a high spore content should be considered as having been formed in the same manner which obtains in present deposits; that is, in open water, and not by an accumulation of fallen plants *in situ*, as stated by the older geological publications upon this subject (**1, 5, 11, 14, 15**).

Since it is generally admitted that natural factors, such as climate and topography, have been instrumental in the formation of our coal beds, it is obvious that a correlation between past and present phenomena is essential for a precise understanding of ancient and modern peat deposits. In regard to climate, competent investigators are quite agreed that there was a somewhat warm and humid atmosphere over the earth during the Carboniferous and later peat-forming epochs. This supposition is corroborated by observations of fossil remains characteristic of the different periods which show a usual lack of annual rings. The nearest parallel to these climatic conditions of growth is now found to prevail only in semitropical and tropical regions. Because of the importance of these considerations, the writer has chosen many of his illustrations from the semitropical peat deposits of Florida, since they present a closer analogy to coal beds than do the more northern organic strata. It has already been pointed out that there is a surprising lack of autocthonous accumulations in this locality in contrast to an abundance of lacustrine deposits. This dearth of land-formed peat is clearly dependent upon the rapid decay of exposed land plants in zones without a winter season. Accelerated disintegration under these conditions is sufficiently pronounced to

prevent any appreciable amassing of vegetable matter other than that protected by a continuous covering of water. Studies of coal sections indicate that similar processes were as effectual in the past, for there is a universal deficiency of strictly autocthonous coals as revealed by the microscope (**8**).

Although this prehistoric lycopod flora, growing on the low-lying shores of ancient lakes, was different from that which now enters into the formation of peat, the process by which fragmentary material was derived from this cryptogamic growth was undoubtedly the same. JEFFREY (**8**), as a result of his studies of sections of coal from all over the world, has found that all categories from cannel to anthracite show spores of arboreal cryptogams in varying amounts, just as the peats of today show different proportions of pollen. In addition to the many spores carried into these carboniferous lagoons by the wind, sluggish streams brought microscopic débris in all stages of decay. This detritus was precipitated, and the allocthonous peat was augmented by an age-long process of sedimentation. A continuance of such conditions finally raised the mass above water when the bordering forests, in zones like the present, marched toward the center and established swamps. This bog-loving flora did not, however, add in any appreciable degree to the substance already accumulated, owing to their rapid decay in a fallen state, both as a result of a warm climate and its less resistant organization. In fact, all microscopical evidence points to a condition very similar to that already described for recent peat deposits, the major part of which is quite conclusively shown to be of drifted derivation.

Another fact which supports the allocthonous theory of coal formation, is the vast predominance of lacustrine peat over *in situ* deposits at the present time. This fact has been well illustrated by the several strata already mentioned, such as those found in open lakes, filled lakes (swamps), and river estuaries. The phenomena are especially noticeable in warm localities, where autocthonous peats are quantitatively almost negligible.

Thus it is apparent that the mode of peat formation, as illustrated by its anatomical structure and topographical features, shows strikingly similar analogies in coal. It must be assumed, therefore,

that the major part of our coal beds, like peat deposits, does not represent a gradual accumulation of successive generations of fallen plants in swamps, but rather a long continued and peaceful sedimentation of wind-blown and drifted plant fragments and minute organisms in the depths of open bodies of water.

In conclusion, the writer wishes to express his sincere thanks to the Committee of Sheldon Traveling Fellowships of Harvard University for the granting of a fellowship, the stipend of which has made possible these investigations; to Professor R. THAXTER of Harvard University for samples of peat; and to Professor E. C. JEFFREY for advice during the course of the work.

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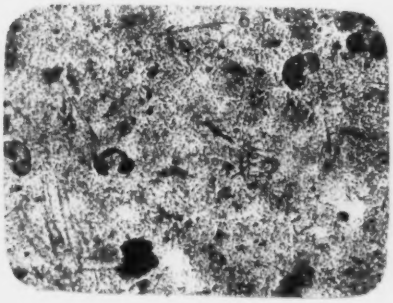

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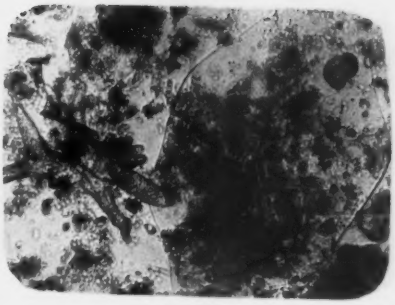

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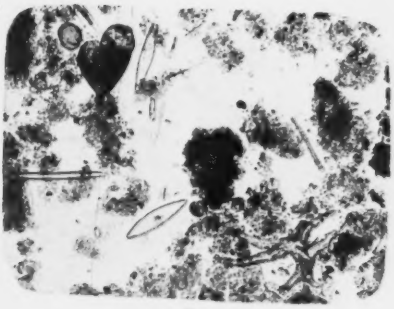

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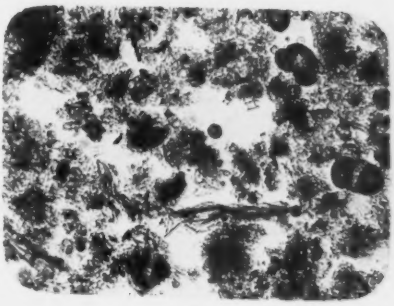

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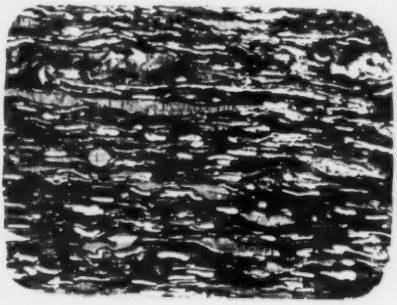

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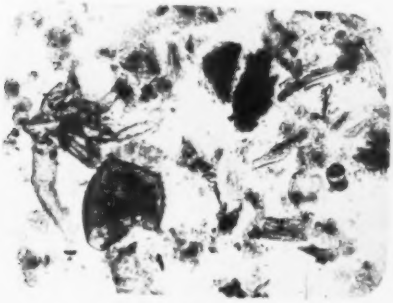
FORSAITH on PEAT



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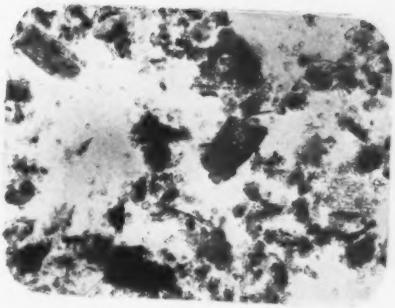

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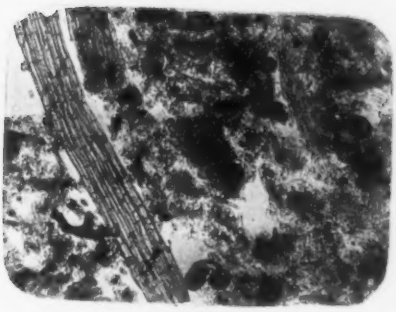

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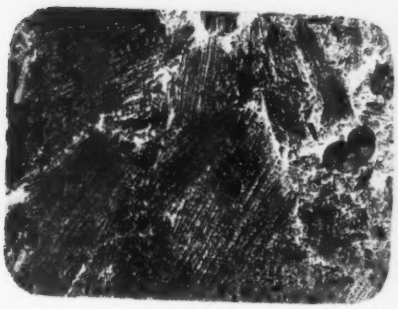

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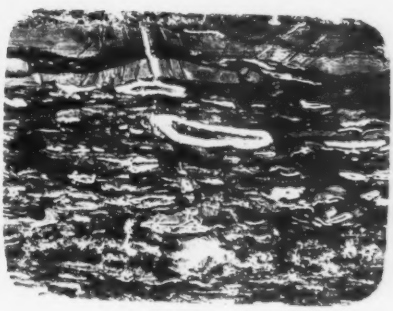

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FORSAITH on PEAT

EXPLANATION OF PLATES X AND XI

PLATE X

FIG. 1.—Composition of allocthonous peat from Lake Harris in Florida; grayish background represents amorphous mass of organic derivation in which there are imbedded abietineous pollen grains, appearing as oblong bodies with 2 laterally attached air sacs; an idioblast from a water lily is pictured in lower left hand corner as a series of spinelike appendages radiating from a common center; other inclusions are dense black ejecta from amphibious animals, and spindle-shaped fresh water sponge spicules.

FIG. 2.—Sample of similar constituents from Lake Dot.

FIG. 3.—Material from Lake Eustis in which there are idioblasts, pollen, spores, spicules, ejecta, and structureless matter; in upper left hand corner there appear 3 specimens of the amoeboid *Arcella;* diatoms of *Stauronesis* and *Navicula* type occur in upper and lower portions of figure respectively.

FIG. 4.—Magnified view of peat from Lake Orange 1 mile off shore; besides characteristic structures, strips of epidermis and an herbaceous plant fragment appear, cells of which are still intact.

FIG. 5.—Organization of Kentucky cannel coal, ×250; scattered throughout the section are numerous light bodies, flattened spores of vascular cryptogams (homologues of the spores and pollen shown in the illustrations of allocthonous peat); the long grayish bands are indicative of metamorphosed bits of wood; separating spores and lignitoid fragments are dense black masses of organic matter.

FIG. 6.—An herbaceous peat from Lake Orange near shore; fragments of roots, etc., manifest cell structure clearly; evidences of drifted material are present, as an idioblast and the mouth part of some insect.

PLATE XI

FIG. 7.—Sample of "saw grass" (*Cladium*) peat 3 ft. from bottom of a marsh bordering Lake Harris; exemplifies the usual inclusions characteristic of allocthonous peat.

FIG. 8.—Preparation of peat 3 ft. from bottom of a bog near Leesburg, Florida, in which are pollen, spores, spicules, ejecta, and other allocthonous inclusions; also woody and herbaceous fragments of plants, cells of which are still intact.

FIG. 9.—Nature of peat above the deep brown plastic layer illustrated in fig. 8; in addition to usual sedimentary matter, intact fragments of herbaceous plants may be seen.

FIG. 10.—Another sample from the same horizontal plane with a much distorted plant fragment in upper right hand corner; below and to left of this, a sponge spicule and fern sporangium may be seen in juxtaposition.

FIG. 11.—A woody peat from Pablo Creek near Jacksonville, Florida, almost entirely composed of lignitoid plant fragments; evidence of a coniferous

origin for this wood is furnished by a uniseriate ray in tangential section and an absence of vessels; indications for a sedimentary origin for this stratum are manifest as a sponge spicule, abietineous pollen, and a group of spores in upper right hand corner.

FIG. 12.—A bituminous coal from Perry County, Ohio, ×250; crinkled bands of compressed wood are especially obvious in upper and left hand parts; evidences of an allocthonous origin for this coal occur as flattened spores, appearing as light bodies imbedded in a dense black amorphous matrix.

THE RESPONSE OF PLANTS TO ILLUMINATING GAS

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 227

SARAH L. DOUBT

(WITH SIX FIGURES)

Introduction

Owing to increasing loss of plants in greenhouses, and of shade trees along city streets, it has seemed worth while to work out simple accurate methods by which gardeners, florists, and foresters may detect gas injury. This study falls into two divisions: (1) injury to greenhouse plants due to presence of gas in the air; and (2) injury to trees and bedding plants due to leakage in the soil. It is hoped that as a result of this work any florist or gardener may be able to determine readily the presence of illuminating gas in the air. The presence of gas in the soil as shown by injury to trees is more difficult to determine.

Considerable work has been done in this laboratory on the effect of illuminating gas and its various constituents upon plants. CROCKER and KNIGHT (1) showed that the buds and flowers of carnations are extremely sensitive to traces of illuminating gas in the air. Three days' exposure, 1 part in 40,000, killed young buds and prevented the opening of those which already showed the petals. The flowers were closed by 12 hours' exposure to 1 part in 80,000. One part ethylene to 1,000,000 parts of air prevented opening of the buds, and 1 part in 2,000,000 caused the flowers to close. Their work showed that ethylene is the constituent which is most toxic to plants. HARVEY and ROSE (**6**) showed that the relatively high toxicity of ethylene holds for many different species of plants. CROCKER, KNIGHT, and ROSE (**2**) found etiolated sweet-pea seedlings to be extremely sensitive to gas, and suggest the use of them as test plants for traces of gas. HARVEY (**8**) suggests the use of the castor bean for the same purpose.

Little is known about the effect of gas on trees. STONE (**9**) has made a number of observations in the field, which led him to

conclude that many trees which are seriously injured by gas finally succumb to attacks of fungi or of insects. He believes these secondary causes are often blamed for the total injury.

Rather striking formative effects have been noted by a number of writers. STONE observed abnormal tissue in the cortex of stems of Carolina poplar, proliferation tissue in the lenticels of willow, and increased root development in cuttings of the willow exposed to gas. HARVEY and ROSE (**6**) found that gas and ethylene cause tubercle-like growths on roots of *Catalpa* and *Ailanthus*.

Methods

The gas used in this investigation was that of the Peoples Gas Light and Coke Company of Chicago. It averages about 4 per cent ethylene and about 25 per cent carbon monoxide. The ethylene used was generated from absolute alcohol and concentrated sulphuric acid and washed in the usual way. It contained 90 per cent ethylene and 10 per cent air impurities. The carbon monoxide was generated from oxalic acid and concentrated sulphuric acid, washed and analyzed. Each of these was mixed with air to give it the same volume percentage as exists in illuminating gas, and the air-gas mixtures were checked against the illuminating gas.

Three general methods of exposing plants to gas were used: (1) the flowers only were treated with gas; (2) potted plants were sealed in wardian cases and measured amounts of gas added; and (3) root systems were treated with a slow stream of gas in the soil.

1. To determine the limit of toxicity for the flower of the calla lily (*Zantedeschia ethiopica*) the method employed by CROCKER and KNIGHT (**1**) upon carnations was used. A 20 liter carboy was inverted over the flower bud and a rubber stopper fitted about the petiole and made gas-tight with vaseline. Gas was forced into this 20 liter bottle through a tube inserted in a second hole in the stopper. A measured amount (800 cc. or 4 per cent) of illuminating gas was forced in, and the pinchcock closed. A control plant was put under identical conditions, except that no gas was used. When the bottles were removed 11 days later, both flowers

opened and were of normal size. The one treated with gas showed slight discoloration on the spathe. Four per cent of illuminating gas, then, slightly injures the inflorescence of the calla lily. Further results on the calla lily are given later.

2. Exposure of entire potted plants was made in two wardian cases (each of about 1000 liters capacity); one case was used for exposure to gas and the other case was used for the control plants. The plants were placed in the cases by the removal of a pane of glass which was later sealed into place with the vaseline-clay mixture used by CROCKER and KNIGHT. The plants were kept under moisture conditions as nearly optimum as possible. During the winter season the temperature varied between 15 and 20° C. with day and night. The experiments were carried on from January to July. In April the temperature became so high in the cases as to injure the plants. The cases were then given a coat of whitewash. As the temperature again rose, the cases were moved outside and partly shaded. Plants were treated for 2 days, then the cases were opened, the plants removed, and the cases aired by means of an electric fan. After watering, the plants were returned to the case and following renewal of gas were left 2 days more. They were then removed and the immediate and the after effect of the gas noted. Since the control plants were in no case injured, it is clear that the response of the other plants was due to the gas.

Types of responses

1. *Leaf fall.*—In certain concentrations of ethylene or of illuminating gas, *Mimosa*, *Lycopersicum*, *Salvia*, *Datura*, *Coleus*, and *Hibiscus* dropped their leaves after a few hours' exposure. The abscission layer was probably formed. The older the plants, the less gas was required to cause the older leaves to drop. The youngest leaves were least affected.

2. *Rigor.*—*Coleus*, *Ricinus*, *Datura*, and *Mimosa* showed rigor when subjected to large amounts of gas. *Mimosa* showed imperfect rigor, lost sensitiveness to touch, but was somewhat injured by the gas. *Coleus* was completely anesthetized, with no ill aftereffects. FITTING (5) found that heat rigor or rigor from lack of oxygen will prevent gas injury to plants.

3. *Epinasty of petioles.*—Suitable concentrations of illuminating gas and of ethylene produced epinasty in petioles or flower stalks. In *Lycopersicum* and *Salvia* this response is often so marked as to produce complete spiral coils. The petioles of *Ricinus*, *Datura*, *Coleus*, and *Hibiscus*, and the flower stalk of calla lily also showed epinasty in traces of these gases. The bending may be near the blade or the bud, as in calla lily leaf and flower; all along the petiole, as in most younger leaves; or very near the stem, as in most older petioles (figs. 1–5).

4. *Proliferation tissue in lenticels, leaf scars, etc.*—In the presence of traces of illuminating gas or of ethylene, soft spongy tissue developed in the lenticels (*Hibiscus* and *Sambucus*), at leaf scars (*Lycopersicum*), or at more or less extensive regions along the stems. In the roots of the apple and pear the abnormal tissue developed just outside the vascular cylinder, but it is not determined whether it was produced by the cortex or the pericycle. Deep longitudinal cracks developed in the bark of the stem. These appeared on the apple, pear, ash, and *Hibiscus*, and to a less degree in *Sambucus*, *Grevillea*, and cottonwood.

5. *Root tubercles.*—Traces of these gases produced tubercle-like growths on the roots of *Grevillea*, *Sambucus*, *Populus*, apple, pear, and *Hibiscus*. In the tomato similar tubercles are produced by nematodes.

Results of treatments

In the following records, plants are arranged in the order of their sensitivity to gas. All amounts of gas indicated are in parts per million of air (ppm).

Lathyrus odoratus.—With 1000, 100, 75, and 50 ppm illuminating gas, the leaves turned yellow and died. Ethylene 8 ppm caused the leaflets to fall off; 5 ppm caused the leaves to become yellow and die; 2 ppm caused death of the older leaves; and 0.1 ppm still caused noticeable injury, although less than in the other cases.

Salvia splendens.—With 25,000 ppm illuminating gas, the older leaves fell off, while the younger ones showed epinasty; 9000 and 8000 ppm caused epinastic response of the petioles;[1] 1000 ppm

[1] In all cases of epinasty the leaves drooped, but the blades and petioles remained rigid. In some cases the halves of the blades folded together somewhat (fig. 4).

caused the oldest leaves at the base of the stem to fall off and the younger leaves showed epinasty; 100, 50, and 25 ppm still caused marked epinasty. With ethylene, 5 ppm caused some leaf fall, epinasty was marked, and some petioles showed a complete spiral coil (figs. 3, 4); 2 ppm caused epinasty but no leaf fall; 0.2 and 0.1 ppm still caused epinasty. With carbon monoxide, 50 and 12.5 ppm caused no response.

1 2

FIGS. 1, 2.—Fig. 1, *Lycopersicum esculentum:* plant at left has been treated for 12 hours with 50 ppm carbon monoxide; plant at right has been treated for 12 hours with 8 ppm ethylene; the former appears perfectly normal, while the latter shows the distinct epinastic response characteristic of gas poisoning; note spiral coiling of one petiole; fig. 2, *Lycopersicum esculentum:* control at left; plant at right has stood for 18 hours in atmosphere containing 1000 ppm illuminating gas; a few hours longer would cause leaf fall, but as they stand the leaves show strong epinastic response; they are bent down, but are stiff.

Mimosa pudica.[2]—With illuminating gas, 60,000 ppm caused imperfect rigor;[3] 100 and 50 ppm caused folded leaflets and pulvinal movement; after a day the leaflets turned yellow and fell off; then some petioles fell; some of the youngest leaves were

[2] With *Mimosa* all amounts of gas used caused the plants to lose their sensitiveness to touch. After recovery they regained it.

[3] The leaflets folded and the leaves drooped as they do at night or after stimulation, but recovery was complete after removal.

uninjured. With ethylene, 8 and 5 ppm caused the same response as the preceding; 2 ppm caused some leaf fall but the injury was less; 0.2 ppm resulted in the fall of a few leaflets, but all leaves showed sensitiveness by folding together; 0.1 ppm caused some leaflets to fall. With carbon monoxide, 50 ppm caused a clear response; leaflets folded and petioles drooped; no leaflets fell; the plant lost its sensitiveness to touch; recovery was complete after two days in air.

Ricinus communis.[4]—With illuminating gas, 60,000 ppm caused imperfect rigor, some leaves falling; 100 ppm caused falling of the older leaves and epinasty of all others; 50 ppm caused marked epinasty but no leaf fall. With ethylene, 8 and 5 ppm caused most of the leaves to fall, the youngest showing epinasty; 2 ppm caused no leaf to fall, but all the leaves showed epinasty; 0.2 and 0.1 ppm caused a less marked response, but epinasty was still evident (fig. 5). With carbon monoxide, 50 and 12.5 ppm caused no response.

Datura Stramonium.—With illuminating gas, 60,000 ppm caused partial rigor; 4000 ppm caused all the leaves except the youngest to fall; 500 ppm caused falling of the oldest leaves; epinasty of the younger leaves was very similar to that of *Ricinus;* 50 ppm caused epinasty of the older leaves. With ethylene, 8 ppm caused the older leaves to fall, the younger leaves showing epinasty; 5 ppm caused less leaf fall, but the remaining leaves showed epinasty; with 2 ppm there was no leaf fall, but evident epinasty; with 0.2 and 0.1 ppm there was evident epinasty. With carbon monoxide, 50 ppm gave no visible response.

Lycopersicum esculentum.—With illuminating gas, 35,000 ppm caused the older leaves to fall, the root growth was stimulated on the stem above the ground, and epinasty occurred;[5] 26,000, 1000,

[4] The epinastic response is very striking in this plant. The cotyledons, leaf blades, and petioles, all show the characteristic turning. The petioles droop about 90° from their normal position, so that instead of making an angle of about 45° with the stem above the leaf, they droop until they make an angle of about 45° with the stem below the leaf. The blades and petioles are rigid after turning, and usually recover their normal position after a couple of weeks with no gas present (fig. 5).

[5] On the older leaves this was near the blade; on the younger leaves it was near the stem. In some cases this growth caused a spiral coil of the petiole (fig. 1).

75, and 50 ppm caused the same kind of response, but the degree was lessened somewhat. With ethylene, 8 and 5 ppm caused fall of the older leaves, the younger leaves showing epinasty (fig. 2), and proliferation tissue developed on the leaf scars; 2 ppm caused a few leaves to fall, but this amount was about the limit for causing leaf fall; 0.2 ppm caused evident epinasty; 0.1 ppm caused no response. With carbon monoxide, 12.5 ppm caused no response.

Coleus sp.—With illuminating gas, 35,000 ppm caused rigor, no leaves fell, and after removal from the cases recovery was com-

3 4

FIGS. 3, 4.—Fig 3, *Salvia splendens:* plant at right was treated with 2 ppm ethylene; after 12 hours it showed distinct epinastic response; plant at left, appearing normal, received 12.5 ppm carbon monoxide; fig. 4, *Salvia splendens:* control plant and one which has been treated for 18 hours in 1000 ppm illuminating gas; epinastic growth of petioles is clear and leaf blades show a folding together of the sides, which is characteristic of presence of considerable gas.

plete; 25,000 ppm caused all the leaves to fall at the end of 24 hours exposure; 6000 ppm caused falling of about half the leaves, the older being the ones affected; 1000 ppm caused the oldest to fall, the younger showed epinasty, and the youngest were unaffected; 100 ppm caused slight epinasty of the younger leaves, and this is close to the limit of response. With ethylene, 5 ppm caused the oldest leaves to fall, while the younger, except those at the tip, showed epinasty; 2 ppm caused no leaf fall, and epinasty was slight; 0.2 ppm caused no response. With carbon monoxide 12.5 ppm caused no response.

Hibiscus rosa-sinensis.—With illuminating gas, 9000, 8000, and 4000 ppm caused all leaves to fall; new leaves developed in 2–3 weeks after removal from the case; the lenticels on the stem were filled with spongy white tissue; with 1000 ppm the older leaves fell, and the younger leaves showed epinasty; with 100 ppm only slight epinasty was evident. With ethylene, 8 ppm caused distinct epinasty; 2 ppm caused slight epinasty, but this is near the limit for response. With carbon monoxide, 12.5 ppm caused no response.

Acalypha tricolor.—With illuminating gas, 25,000 ppm caused some leaf fall, and other leaves showed epinasty; 8000 and 1000 ppm caused no leaf fall, but distinct epinasty.

Acacia horrida.—With illuminating gas, 8000 ppm caused fall of many leaves; 1000 ppm caused fall of some leaves several days after treatment.

Euonymus japonicus.—With illuminating gas, 20,000 ppm caused most of the leaves to fall; 8000 ppm caused the older leaves to fall; 1000 ppm caused a few of the older leaves to fall.

Citrus decumana.—With illuminating gas, 20,000 and 8000 ppm caused most of the leaves to fall after removal from the case; 1000 ppm caused the older leaves to fall.

Zantedeschia ethiopica.—With illuminating gas, 40,000 ppm caused the flower spathe to become somewhat discolored, but the plant seemed uninjured; 10,000 ppm caused epinasty in the young leaves, the petioles being arched next to the blade; 9000, 8000, and 1000 ppm caused the youngest leaf and the peduncle to show epinasty as above. With ethylene, 5 ppm caused slight epinasty of the youngest leaf; 2 ppm caused no response.

Pelargonium zonale.—With illuminating gas, 25,000 and 4,000 ppm caused no visible response during 4 days of treatment, but all leaves fell in 3–6 days after treatment had stopped;[6] 100 ppm caused no response. With ethylene, 8 and 2 ppm caused no response. With carbon monoxide, 12.5 ppm caused no response.

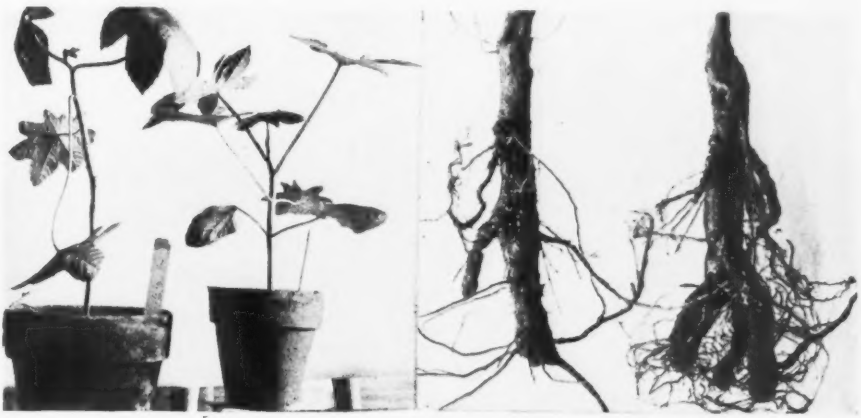
Begonia luminosa.—With ethylene, 8 ppm caused some epinasty at the base of the leaf blade. With carbon monoxide, 50 ppm caused no response.

[6] When new leaves developed, they were without the variegated zone.

Fuchsia speciosa.—With illuminating gas, 20,000 ppm caused some of the older leaves to fall; 8000 ppm caused epinasty.

Populus deltoides.[7]—With illuminating gas, 35,000, 25,000, and 10,000 ppm caused some leaf fall, and after removal from the cases other leaves died and fell off; 8000 ppm caused some leaf fall.

Ficus elastica.—With illuminating gas, 20,000 ppm caused the older leaves to fall during treatment; 8000 ppm caused some of the older leaves to fall about a week after treatment.



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FIGS. 5, 6.—Fig. 5, *Ricinus communis:* plant at right, appearing normal, stood for 2 days in 50 ppm carbon monoxide; plant at left stood in 8 ppm ethylene for 2 days, and has dropped one leaf, all other petioles showing strong epinastic response; note that direction of leaf blades is altered as well as that of petioles; fig. 6, Roots (*Pyrus communis* at right; *P. Malus* at left): the 3-pronged root of pear has had 100 liters of illuminating gas forced into the soil during 40 days; notice swollen condition of underground parts, also numerous "tubercles"; the root of apple received 160 liters of illuminating gas in 62 days, its response, swelling and tubercles, being very similar to that of pear.

Croton tiglium var. Sanders.—With illuminating gas, 20,000 ppm produced some leaf fall of the older leaves; 8000 ppm caused slight epinasty.

Tulipa (several varieties).—With illuminating gas, 10,000 ppm caused injury of the flower buds and the tips of the younger leaves rolled up; 4000 ppm caused no visible injury.

[7] These were cuttings rooted in sand and grown in flower pots.

Hyacinthus (several varieties).—With the same quantities of illuminating gas, the responses were identical with those of the tulip.

Carica Papaya.—With illuminating gas, 20,000 ppm caused the older leaves to fall and the younger leaves to show epinasty.

Caladium esculentum.—This showed no response with 75 ppm illuminating gas, 8 ppm ethylene, or 50 ppm carbon monoxide.

Lupinus perennis.—This showed no response with 8 ppm ethylene or 50 ppm carbon monoxide.

Eriobotrya japonica, *Phoenix canariensis*, *Conocephalus* sp., *Canna* (King Humbert and other varieties), *Achyranthes Lindini*, *Cytisus canariensis*, and *Alternanthera* sp. showed no response with 20,000 ppm illuminating gas.

Polypodium, *Aspidium*, and *Asplenium.*—With illuminating gas, 60,000, 8000, and 4000 ppm caused no response.

The preceding data are summarized briefly in table I. The plants are grouped according to their sensitiveness to gas: very sensitive, less sensitive, and resistant. The minimum concentration necessary to produce a response is given in each case.

The following plants showed no response to illuminating gas or to ethylene in the concentration used: *Caladium esculentum*, *Lupinus perennis*, *Eriobotrya japonica*, *Phoenix canariensis*, *Conocephalus*, sp., *Canna*, *Achyranthes Lindini*, *Alternanthera* sp., *Cytisus canariensis Polypodium*, *Aspidium*, and *Asplenium.*

Root treatment of trees

Two or three year old trees were used for the root treatment. They were treated in flower pots during the winter and early spring, and then the work was carried on out of doors upon young trees which had been growing in the soil for a year or more.

The potted plants were set on tripods and glass tubing was run through the cork plug in the bottom of the pot. Connection was made with a wash bottle and the rate of gas flow through this wash bottle was controlled by means of a brass stopcock. The gas was forced out from the inverted carboys by means of water from a raised tank. All rubber connections with glass tubing were as short as possible, gas tight, and the gas was "water sealed" in

the inverted carboys. By means of the brass stopcocks and glass tubes drawn out to a fine point in the wash bottles, the rate of flow of the gas could be regulated at will. To keep the soil from plug-

TABLE I

Plant	Gas used and amount in parts per million (ppm)	Response
VERY SENSITIVE PLANTS		
Lathyrus odoratus	Illuminating gas, 25	Leaflets died and fell off
Salvia splendens	Illuminating gas, 25	All leaves showed epinasty
Mimosa pudica	Illuminating gas, 50	Some leaflets fell; others showed epinasty
Ricinus communis	Illuminating gas, 50	Epinasty shown by the leaves
Datura Stramonium	Illuminating gas, 50	Epinasty shown by the younger leaves
" "	Ethylene, 0.1	Close to the limit for the response
Lycopersicum esculentum	Illuminating gas, 50	Epinasty
" "	Ethylene, 0.2	Epinasty shown by the leaves
" "	Ethylene, 0.1	No response
LESS SENSITIVE PLANTS		
Coleus sp.	Illuminating gas, 100	Slight epinasty shown by the younger leaves
Hibiscus rosa-sinensis	Illuminating gas, 100	Epinasty shown by the older leaves
Acalypha tricolor	Illuminating gas, 1000	Epinasty
Acacia horrida	Illuminating gas, 1000	Some leaves fell
Euonymus japonicus	Illuminating gas, 1000	Some leaves fell
Citrus decumana	Illuminating gas, 1000	Some leaves fell
Zantedeschia ethiopica	Illuminating gas, 1000	Youngest leaf arched at the base of the blade
Pelargonium zonale	Illuminating gas, 4000	Leaves fell several days after treatment
Begonia luminosa	Ethylene, 8	Slight epinasty
Fuchsia speciosa	Illuminating gas, 8000	Epinasty
Populus deltoides	Illuminating gas, 8000	Some leaves fell
Ficus elastica	Illuminating gas, 8000	Some leaves fell
Croton tiglium	Illuminating gas, 8000	Slight epinasty
Tulipa (several vars.)	Illuminating gas, 10,000	Flower buds somewhat injured
Hyacinthus (several vars.)	Illuminating gas, 10,000	Flower buds somewhat injured
Carica Papaya	Illuminating gas, 10,000	Older leaves fell, the younger showed epinasty.

ging the glass tube inserted in the pot, a small vial with a slit along the side of the cork was fitted over the end of the tube inside the flower pot. The pot was then dipped into melted paraffin in order to prevent too much escape of gas through the lower part of the pot.

When the trees were treated in the open ground, glass tubes 12–24 inches long, depending upon the size of the trees, were buried close to the side of the tree. The same precaution was used here to prevent clogging.

The following are the results for each tree or plant treated, the length of time treated, and the amount of illuminating gas passed into the soil. In no case could the odor of the gas be detected on a handful of the soil or escaping in the air. The temperature range was 12–20° C.

Acer Negundo.—A young tree was treated for 45 days and given 60 liters of gas. There was no visible effect above or below ground.

Acer saccharinum.—Treated 42 days and given 140 liters of gas, the parts above ground were unchanged. The stem below ground, however, was swollen, soft, and cracked longitudinally. A section showed proliferation tissue produced just outside the vascular cylinder.

Chrysanthemum hortorum.—One plant, being treated 42 days and given 80 liters of gas, was killed, no proliferation tissue being produced or other visible changes. A second plant was treated 28 days and given 60 liters of gas. Some roots grew up out of the ground, probably due to loss of geotropic response.

Fraxinus americana.—Treated for 42 days and given 120 liters of gas, the parts above ground were unchanged. Below ground the stem was swollen, soft, and cracked longitudinally. Sections showed that abundant proliferation tissue was produced just outside the vascular system.

Grevillea robusta.—One specimen was treated 33 days and given 40 liters of gas. After 2 weeks gummy matter exuded from a slight crack in the stem just above the ground. A second plant was treated 48 days and given 40 liters of gas; and a third was treated 31 days and given 19 liters of gas. The roots of all three showed spongy white masses of tissue at short intervals, with no epidermis. Many roots were dead. The underground parts of the stem were swollen, due to the development of spongy, white tissue.

Hibiscus rosa-sinensis.—A plant was treated 15 days and given 40 liters of gas. The leaves showed epinasty for 2 days and then fell off. After 4 days' treatment, white spongy tissue developed

in the lenticels just above ground. The underground parts were enlarged to three times their normal size. The cortical tissue was white and spongy. The bark split longitudinally and dropped off. Small tubercles developed on many roots. The xylem and phloem appeared normal. These results with *Hibiscus* agree with those of HARVEY and ROSE (**6**).

Lycopersicum esculentum.—One plant was treated 24 days and given 20 liters of gas, while a second plant was treated 18 days and given 20 liters of gas. After a few hours' treatment, the lower leaves began to show the epinastic response, falling after 2 days. Many more roots developed on the stem above ground than on the control plant. Roots grew up out of the ground, probably due to loss of geotropic sensitiveness. Tubercles occurred on the roots. The control plants showed some tubercles, but the number was greatly increased upon the treated plants. Nematodes were present in many of these tubercles.

Poa pratensis.—One sod was treated 38 days and given 60 liters of gas; a second 25 days with 60 liters; a third 5 days with 40 liters; and a fourth 8 days with 40 liters. There was no response in any case.

Populus deltoides.—Treated 81 days and given 60 liters of gas, the roots developed many small "tubercles," being swollen to twice the normal size at these points. The tissue was soft and spongy. The stem showed no visible effect above ground; below ground it was swollen and rigid.

Pyrus communis.—Treated 40 days and given 100 liters of gas, there was no visible response above ground, but all underground parts were swollen. Longitudinal cracks appeared, in which was soft spongy tissue. All the roots were irregularly enlarged, all the proliferation tissue being in the cortex (fig. 6).

Pyrus Malus.—Treated 62 days and given 160 liters of gas, the response was very similar to that of the pear (fig. 6).

Ricinus communis.—Treated 45 days and given 80 liters of gas, all leaves except the youngest fell. The underground part of the stem was swollen and cracked longitudinally.

Salvia splendens.—One plant was treated 18 days and given 20 liters of gas, and another plant was treated 42 days and given

80 liters of gas. Some leaves fell and others showed epinasty; but the underground parts showed no effect.

Sambucus canadensis.—Treated 60 days and given 60 liters of gas, the roots were killed. Some roots which were still living showed "tubercles" similar to those upon *Populus*. The stem below ground was somewhat swollen, due to the development of spongy white tissue in the lenticels.

Ulmus americana.—Treated 90 days and given 180 liters of gas, after 3 weeks the bark cracked vertically just above the surface of the ground. After 6 weeks, 2 small limbs died and were removed. About half the leaves fell during the treatment. Near the close of the treatment the underground parts were dead and cracks extended throughout the bark and cortical tissue. There was a small amount of proliferation tissue just outside the vascular system.

Practical suggestions for florists

To detect illuminating gas in a greenhouse, the florist should provide himself with some vigorous plants of one of the following: tomato, castor bean, scarlet sage, Jimson weed, or sensitive plant. These should be grown in pots so that they may readily be handled, and should have from 6 to 12 or more leaves. They must also be in vigorous condition; otherwise they may not respond should illuminating gas be present. These should be placed at various locations throughout the greenhouse and left 24–48 hours with poor ventilation. All the plants named will respond to traces of illuminating gas within this period at ordinary temperatures.

With only a trace of gas present in the air, the epinastic response of the leaves will be very noticeable if these plants are compared with normal plants without gas. This bending down of the leaves will increase with the concentration of the gas present in the air. All these plants will drop their leaves with a concentration below the limit of the odor of gas. The older leaves fall first, the younger leaves being retained until there is 1 part of illuminating gas to 1000 of air.

Summary

1. The following plants are admirably adapted for use as test plants for illuminating gas in greenhouses: *Lycopersicum escu-*

lentum, *Salvia splendens*, *Mimosa pudica*, *Ricinus communis*, *Datura Stramonium*, and *Dianthus Caryophyllus*. The response of each is definite, striking, and not easily mistaken.

2. Traces of gas (50 ppm of air) cause the epinastic growth of the petioles of all these plants, with the exception of the last. The flower buds of the carnations are blighted by these amounts. One part of illuminating gas per 1000 of air causes leaf fall in the following plants: *Lycopersicum esculentum*, *Salvia splendens*, *Mimosa pudica*, *Datura Stramonium*, *Ricinus communis*, *Coleus* sp., and *Hibiscus rosa-sinensis*. Both the amounts (50 ppm of air and 1 part per 1000 of air) are far below the limit of odor. Repeated trials showed that it was impossible to detect less than 0.25 per cent of illuminating gas (1 part to 400 of air) by the sense of smell.

3. Amounts of ethylene corresponding to the gas mixture gave similar responses; 2 ppm of air caused epinastic growth of the petioles of *Lycopersicum esculentum*, *Salvia splendens*, *Mimosa pudica*, *Ricinus communis*, and *Datura Stramonium;* 8 ppm of air (equivalent to 200 parts of illuminating gas) caused some leaf fall in the 5 plants named.

4. *Poa pratensis* and *Acer Negundo* are very resistant to gas, having shown no response to concentrations injurious to all other forms tested.

5. The following plants are not noticeably injured by gas unless there is enough present to be detected by odor: *Caladium esculentum*, *Lupinus perennis*, *Eriobotrya japonica*, *Phoenix canariensis*, *Conocephalus* sp., *Canna*, *Achyranthes lindini*, *Alternanthera* sp., *Cytisus canariensis*, *Polypodium*, etc.

6. The following trees are rather sensitive to gas escaping into the soil: *Pyrus Malus*, *P. communis*, *Fraxinus americana*, *Ulmus americana*, *Sambucus canadensis*, *Grevillea robusta*, *Catalpa speciosa*, *Populus deltoides*, and *Tilia americana*. Apple, pear, ash, elm, *Catalpa*, and *Sambucus* showed proliferation tissue in the cortex of the stems below the surface of the ground. Elm, ash, and cottonwood showed longitudinal cracks in the bark just above the surface of the ground.

7. The following bedding plants are injured by gas escaping into the soil: *Lycopersicum esculentum*, *Salvia splendens*, *Ricinus communis*, and *Chrysanthemum hortorum*. *Chrysanthemum* is

killed outright; the others drop their leaves or show epinastic growth of the petioles.

8. Young trees at least are injured by leakage of illuminating gas too slight to be detected by odor. The foliage shows no injury, and one would not be likely to suspect gas poisoning from the appearance of the tree above ground. Judging from my results with trees, their killing by illuminating gas is a very slow process, going on for months or years. It is certain that enough gas to cause an odor in the vicinity of trees would be enough to injure them seriously.

I am indebted to Dr. WILLIAM CROCKER for suggestions and help during the progress of the work.

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THE SUPPOSED ACTION OF POTASSIUM PERMANGANATE WITH PLANT PEROXIDASES

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REED[1] has recently reported experiments which he believes throw a new light on the mechanism of oxidation in living tissues. The experiments relate to the reactions involved in the process of oxidation by means of peroxidases. To a horseradish extract, which in itself was incapable of bringing about the oxidation of potassium iodide or of gum guaiac, he added concentrated potassium permanganate solution until the permanganate was no longer reduced. He then added a small excess of horseradish extract to reduce any free permanganate present. On filtration a clear, rather deep yellow solution was obtained, which, when mixed with solutions of potassium iodide, gum guaiac, or pyrogallol, caused rapid oxidations of those substances. REED's interpretation of these experiments is that the peroxidase of the horseradish extract combines with oxygen from the permanganate, thus forming a new compound which readily gives up oxygen to other compounds. He concludes, therefore, that in oxidation processes catalyzed by peroxidases two reactions are involved: first, a combination of the peroxidase with oxygen from substances acting as oxygenases; and second, a transfer of this oxygen to the substances oxidized by means of peroxidases. Thus he believes the mechanism of oxidation in living tissues is explained.

Contrary to REED's belief, this interpretation throws no new light on the mechanism of oxidation in living tissues, but is quite in harmony with the older views of TRAUBE, BACH, ENGLER, and KASTLE and LOEVENHART. Moreover, since manganese compounds themselves are capable of bringing about oxidations, the correctness of REED's view ascribing the oxidations in the mixtures with which he worked to peroxidase activated by oxygen from the postassium permanganate is at least open to question. That the

[1] REED, G. B., The mode of action of plant peroxidases. BOT. GAZ. 62:233–238. 1916.

presence of peroxidases is not necessary to bring about the observed reactions is shown by the following experiments:

1. Two-tenths of a gram of dried white of egg was dissolved in 10 cc. of water. To this solution 2 drops of saturated solution of potassium permanganate were added. The pale brown filtrate from this mixture gave intense oxidase reactions with guaiac, potassium-iodide-starch, and pyrogallol.

2. One gram of Witte's peptone was dissolved in 20 cc. of water and 5 drops of saturated potassium permanganate solution were added. With the pale brown filtrate the 3 oxidase reactions just mentioned were carried out. All were strongly positive.

3. To about half a gram of tyrosin mixed with water 2 drops of a 5 per cent solution of potassium permanganate were added. The brownish mixture gave a clear brown filtrate which oxidized potassium iodide, guaiac, and pyrogallol.

4. One-half gram of tyrosin was dissolved in hot water. To the boiling solution 3 drops of a 5 per cent solution of potassium permanganate were added. The pale brown filtrate from the mixture gave the 3 oxidase reactions. This filtrate was boiled and allowed to stand overnight, but no further precipitate was formed. The filtrate still gave all the oxidase reactions.

5. To a solution of 1 gram of glucose, 2 drops of a saturated potassium permanganate solution were added. The mixture was warmed until the purple color had given way to light brown. The filtrate gave the oxidase reactions distinctly. Fructose treated in the same way gave strong reactions with guaiac and with potassium-iodide-starch, but none with pyrogallol.

6. To a boiling solution containing 3 drops of a 5 per cent potassium permanganate solution in 10 cc. of water, glycerine was added drop by drop until the purple was replaced by brown. The filtrate from the brown precipitate was pale straw-colored. It gave all the oxidase reactions.

7. One cc. of salicylic aldehyde reduced quickly in the cold 2 drops of a 5 per cent solution of potassium permanganate, forming a brown precipitate. The pale straw-colored filtrate gave reactions with potassium-iodide-starch and with guaiac.

8. Methyl alcohol and ethyl alcohol when gently warmed reduced potassium permanganate to a colorless solution, which when filtered from the brown precipitate gave no oxidase reactions.

9. Formaldehyde and acetaldehyde reduced potassium permanganate in the cold, giving brown or black precipitates and colorless solutions which gave no oxidase reactions. This result was to be expected from the action of methyl alcohol and ethyl alcohol.

10. A trace of manganese peroxide shaken up in water oxidized guaiac, potassium iodide (in neutral solution), and pyrogallol.

In all these experiments, a large excess of the organic compounds was used, so that the solutions would be free from potassium permanganate in the sense in which REED considered his solutions free from unreduced potassium permanganate. The potassium iodide reactions were carried out in solutions of the same strength as those used by REED. In all cases the reagents and control mixtures failed to give the oxidase reactions. The brown filtrates as well as the colorless ones contained manganese.

It appears from these experiments that in the reduction of potassium permanganate by organic substances in neutral solutions, hydrated peroxides of manganese are formed, which are held in solution and which though they are reduction products of permanganic acid are still capable of carrying out oxidations. When the permanganate is still further reduced, the water-clear filtrate which still contains manganese compounds no longer gives the oxidation reactions described. By careful reduction both stages can be obtained with the same compound (glycerine, glucose).

Inasmuch as the brown solutions contain manganese not reduced to its lowest state of oxidation, and since manganese peroxide itself brings about the oxidation of guaiac, potassium iodide, and pyrogallol, it becomes extremely probable that the oxidation phenomena observed by REED were brought about by peroxides of manganese and not by activated plant peroxidases. Moreover, since a number of substances acting on potassium permanganate give mixtures which oxidize other compounds, there is no evidence in REED'S experiments that the reduction of the

potassium permanganate was brought about by plant peroxidases. His conclusions, therefore, drawn from reactions which are common to many organic substances and which are not known to be properties of peroxidases, are too sweeping for the experimental grounds upon which they are based.

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LEAF NECTARIES OF GOSSYPIUM

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(WITH PLATES XII AND XIII AND ONE FIGURE)

On the midrib and sometimes on the other principal veins on the underside of leaves of *Gossypium*, certain nectar glands are found. All species of cotton, with the possible exception of *G. tomentosum*,

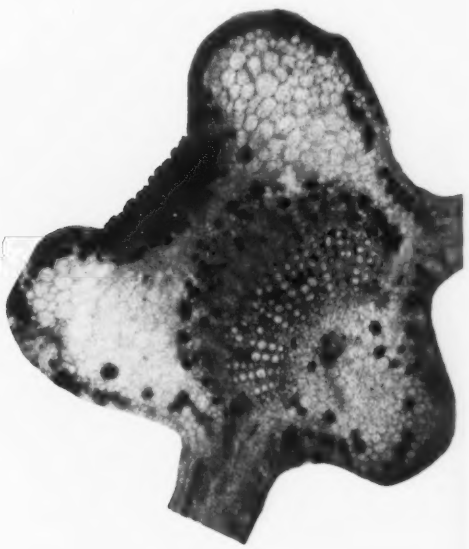


FIG. 1.—Nectar gland on a leaf of *Gossypium hirsutum*

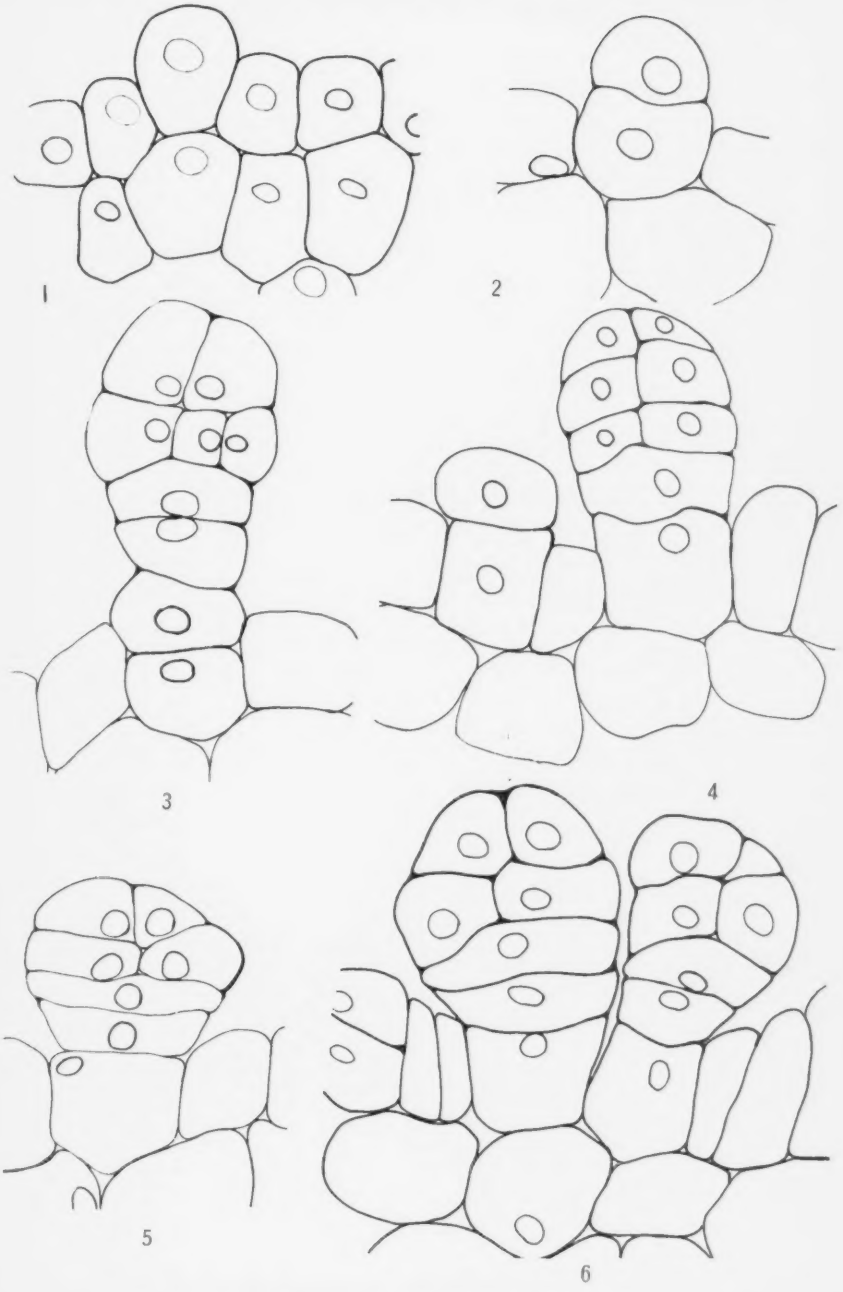
possess these glands, which are usually oval or pear-shaped, sometimes even sagittate in form. TAYLOR (1) says "these glands are usually small, rounded, shallow pits, with a floor of round-topped secreting cells"; and WATT (2) states that "these midrib glands may be elongated and elevated portions of the veins that

become pale colored or assume a pink tinge, and then rupture lengthwise, or they may be circular or oblong warts which open into distinct pits." TRELEASE (3) points out that the glands begin to secrete at about the time the seedling has expanded 4 leaves, and that the nectar is secreted most abundantly at night. SAFFORD (4) states that "they . . . . occur on all leaves of cotton . . . . in the form of vaginated glands." He gives a photograph by HOWARD, of the United States Department of Agriculture, of a cross-section of a nectar gland of the cotton leaf.

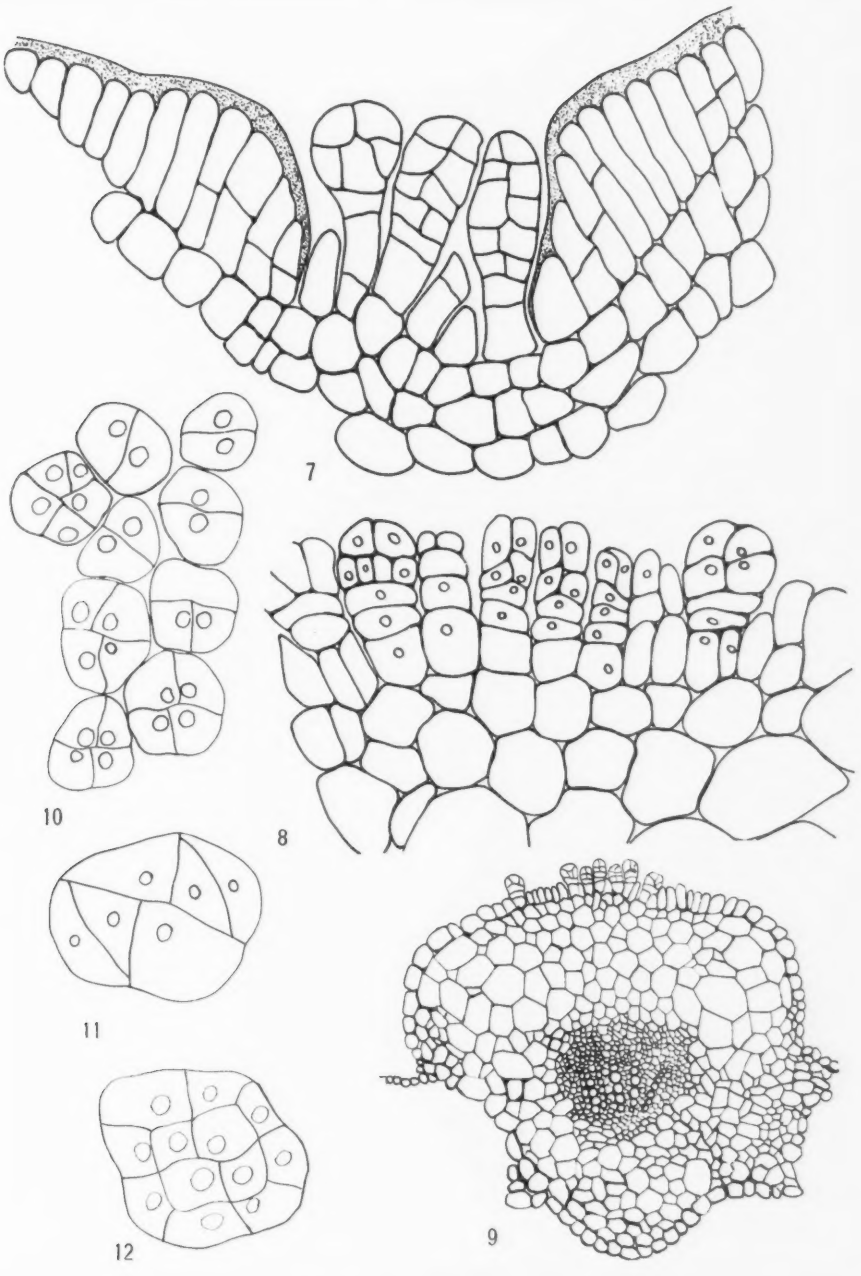
The glands described in this paper are from *Gossypium hirsutum*. They are oval-shaped depressions, filled with closely crowded multicellular papillae and surrounded by a thick wall of epidermal cells (text fig. 1 and fig. 7). In all cases observed the glands became visible on the cotyledons about the time the first pair of true leaves developed; they began to secrete a little later. Sections were made of the cotyledons as soon as they were fully expanded; of the second and third pair of true leaves at different stages of development; and also of mature leaves. A section through a gland of a mature leaf is shown in fig. 7 and one through that of a young leaf in fig. 9. These glands are of epidermal origin and consist of numerous multicellular papillae (text fig. 1). Their organogeny is as follows:

The epidermal cells from which the glands arise cease to develop normally and become papillate (fig. 1). The papillae are next cut off by transverse walls (fig. 2). The cells thus formed divide again in the same plane; this may be repeated once or twice, and results in the formation of short pedestals consisting of 2, 3, or 4 cells (figs. 3–6). The terminal cell of each papilla then divides by a vertical wall into two (fig. 10). These in turn divide by walls at right angles to the first cross wall into 4 cells (fig. 10). The latter divide by periclinal walls into 4 central and 4 peripheral cells (fig. 11). Lastly, each of the external cells divides by a wall at right angles to the surface, and thus a peripheral layer of 8 cells is formed (fig. 12). The development of the papillae of these glands bears a remarkable resemblance to that of the antheridia of *Riccia* (5).

COLLEGE STATION
TEXAS



REED on LEAF NECTARIES



REED on LEAF NECTARIES

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2. WATT, GEORGE, Wild and cultivated cotton plants of the world.
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4. SAFFORD, W. EDWIN, Useful plants of Guiam.
5. CAMPBELL, D. H., Mosses and ferns.

EXPLANATION OF PLATES XII AND XIII

PLATE XII

FIG. 1.—From transverse section of leaf showing papillate cell.

FIG. 2.—First cell of pedestal cut off by cross wall.

FIGS. 3, 4, 5, 6.—Multicellular papillae showing pedestals with varying number of cells.

PLATE XIII

FIG. 7.—Vertical section through gland near one end showing papillae and elongated cells; some of the latter have divided horizontally, and have formed the wall of the gland.

FIG. 8.—Vertical section through young gland showing first stage in formation of wall (right side of figure).

FIG. 9.—Vertical section through midrib showing young papillae.

FIG. 10.—Cross-section of several papillae showing first and second vertical division.

FIG. 11.—Cross-section of papilla showing periclinal walls.

FIG. 12.—Cross-section of mature papilla showing divisions subsequent to those shown in fig. 11.

THE REACTION OF PLANT PROTOPLASM

A. R. HAAS

The reaction of protoplasm is one of the most important factors of metabolism. It is not, however, the apparent reaction (or total acidity), as shown by titration, which is of chief importance, but the actual reaction, as shown by the gas chain or by indicators. The total acidity includes both undissociated and dissociated acid, while the actual reaction depends only upon the latter.

In the case of a buffer solution[1] the total acidity may be very high, while the actual acidity may be very low. The higher the total acidity in this case the more difficult it becomes to change the actual reaction by the addition of acid or alkali. This applies to protoplasm, which always has the properties of a buffer solution (since it contains carbonates, phosphates, proteins, etc.). Hence if a low actual acidity is advantageous to the protoplasm, a high total acidity may be favorable, in that it preserves this desirable actual acidity.

So far as the writer is aware, no determinations of the actual reaction of plant protoplasm have been made by means of the gas chain (except a single determination of pineapple juice made by REED[2]). Only a few determinations have ever been made[3] by means of indicators.

The writer has investigated the reaction of a number of plants, including some which seemed to be of special interest on account of their unusual acidity. The method employed was to crush the cells and to determine the acidity of the juice by means of the gas chain. The gas chain used was essentially the form described by HILDEBRAND.[4]

[1] Cf. HÖBER, H., Physikalische Chemie der Zelle und der Gewebe (pp. 118, 169). Berlin. 1914.

[2] Unpublished results.

[3] Cf. FRIEDENTHAL, H., Zeit. Allg. Physiol. **1**:56. 1902. It is not known whether in these experiments the proper precautions were taken to crush all the cells and to secure plant juices which had as high an acidity as the cell contents.

[4] HILDEBRAND, J. H., Jour. Amer. Chem. Soc. **35**:869. 1913.

The method of obtaining the juice is of considerable importance. It has been shown by MAMELI,[5] by MARIE and GATIN,[6] and by DIXON and ATKINS,[7] that when tissue is crushed the juice which is first expressed contains a much lower concentration of electrolytes than that which is obtained when greater pressure is applied so as to crush more of the cells. It is obvious that when pressure is first applied and sap is squeezed out through the intact plasma membrane, the electrolytes may largely be retained within the cell because they are not able to pass freely through the membrane. It is desirable, therefore, to grind the tissue and rupture all of the cells. This was accomplished by thoroughly grinding the tissue in a mortar. Only a little tissue was ground at a time, and the grinding was continued until microscopic examination showed that all the cells were ruptured.

TABLE I

Material	Actual acidity of CO_2-free undiluted juice as determined by the gas chain	Total acidity as determined by ordinary titration methods
Lemon (fruit)	0.006N	
Cranberries (fruit), fresh material, peeled and unpeeled	0.004N	0.9172N (ripe fruit) 0.3104N (overripe, soft fruit) 0.3403N (overripe, soft fruit)
Grapefruit (fruit)	0.001N	0.1927N
Apple (fruit)	0.0004N	0.0711N
Rhubarb (leaf-stalk) basal part	0.0007N	
Rhubarb (leaf-stalk) intermediate part	0.0005N	0.1578N
Rhubarb (leaf-stalk) green part below leaf blade	0.00022N	0.1681N
Orange (fruit)	0.00016N	0.0941N
Pineapple (ripe fruit), juice standing 2 hours	0.00009N	
Pineapple (ripe fruit), fresh juice	0.000035N	0.1377N
Green pepper (fruit)	0.000038N	
Eggplant (fruit)	0.000002N	

Before determining the acidity of the juice, the CO_2 was driven off by means of a current of hydrogen. The results of the determinations are given in table I. The results in all cases represent the average of two or more closely concordant determinations.

[5] MAMELI, E., Atti Ist. Bot. Univ. Pavia **12**:285. 1908.

[6] MARIE, C. H., and GATIN, C. L., Déterminations cryoscopiques effectuées sur des sucs végétaux. 1912.

[7] DIXON, H. H., and ATKINS, W. R. G., Proc. Roy. Dublin Soc. **13**:422. 1913.

The figures of the table show that the actual acidity bears no fixed relation to the total acidity, and that great variations are to be found in different plants, as well as in different tissues of the same plant.

The figures for actual acidity are surprisingly high in the case of the lemon and of the cranberry, especially in view of the prevalent opinion that protoplasm demands a neutral or nearly neutral reaction for normal metabolism. It is of interest, therefore, to inquire whether the figures represent the actual acidity of the protoplasm. In the lemon the acid juice is contained in sacs, the walls of which are composed of living cells, while the cavity is produced by the disintegration of cells. REED[8] has shown that the living cells of the walls contain oxidases whose activity is promptly inhibited by the acid contained in the cavity of the sac. Since the oxidases are active in the living cells, it follows that the protoplasm is by no means as acid as the juice in the sacs, and hence the figures given in table I cannot apply to the protoplasm in the case of the lemon.

With the cranberry the case seems to be different. In this fruit there are no sacs such as are found in the lemon; the juice is contained entirely in the cells. It is important, therefore, to ascertain whether these cells are dead or alive. In order to test this, the outer colored layer of cells was removed and the following results were obtained on the colorless cells:

1. Eosin failed to penetrate unboiled peeled cranberries, but penetrated readily into the boiled peeled cranberries. The difficulty due to the precipitation of eosin in acid solutions was obviated by frequent renewal of the eosin solution.

2. Ripe cranberries can be peeled without staining the white tissue beneath, while this is not possible in overripe, soft cranberries.

3. Unboiled, peeled cranberries in a red watery extract of cranberry peeling were unstained after several hours, while the indicator readily penetrated the boiled peeled cranberries.

All these tests go to show that the living cells of the cranberry have an actual acidity which is extremely high. It is quite pos-

[8] REED, G. B., BOT. GAZ. 57:528. 1914.

sible, however, that the acid sap is contained in vacuoles rather than imbibed in the protoplasm proper.

Summary

1. The actual acidity and the total acidity of a number of plant tissues were determined.

2. There is no constant relation between the two, but great variations occur in different plants and in different parts of the same plant.

3. In one case (cranberry fruits) the surprisingly high actual acidity of 0.004N (as determined by the gas chain) was found in the living cells.

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BRIEFER ARTICLES

A FREEZING DEVICE FOR THE ROTARY MICROTOME

(WITH ONE FIGURE)

A few years ago OSTERHOUT[1] figured and published an account of a simple freezing device to be used in connection with various sorts of sliding microtomes. Later[2] he published an account of a simple freezing microtome in which he made use of a knife of a plane for cutting on account of its rigidity. In each of these devices, the freezing chambers being stationary, they are both adaptable to the use of brine, carbon dioxide, or other substances for freezing. These devices have been of considerable service in the preparation of sections of living tissues. However, if one wishes sections in large quantity and of uniform thickness, and particularly if thin sections are desired, it is found that a sliding microtome of almost any construction is inadequate, and to manipulate it requires considerable dexterity.

It occurred to the writer that the OSTERHOUT apparatus for freezing with brine might be modified in such a way as to make it usable with a rotary microtome of any make, and thereby increase its efficiency and enlarge the usefulness of both pieces of apparatus. The adaptation was made and the results have proven so satisfactory that a brief account of the apparatus seems desirable.

The accompanying photograph of the apparatus will serve as a basis for the description. It is simple and easy to construct, consisting of a 2×10 board 3.5 ft. long for the base, and 2 upright pieces fastened at the base and braced by a cross-piece about one-third of the distance up. A bolt passes through the base and the center of the cross-piece, and another through the upright pieces just above the cross-piece to make the apparatus firm. The upright has been lengthened in this case to receive larger receptacles than were originally used. The wheel is 20 inches in diameter and 1.5 inches thick, making the whole device about 5 ft. high. The wire to hold the pails is firmly fastened in the middle to the grooved wheel. The rubber tubing is of stiff white rubber. When

[1] OSTERHOUT, W. J. V., A simple freezing device. BOT. GAZ. **21**:195–201. *figs.* 6. 1896.

[2] ———, Univ. Calif. Publ. Bot. **2**:73. 1904.

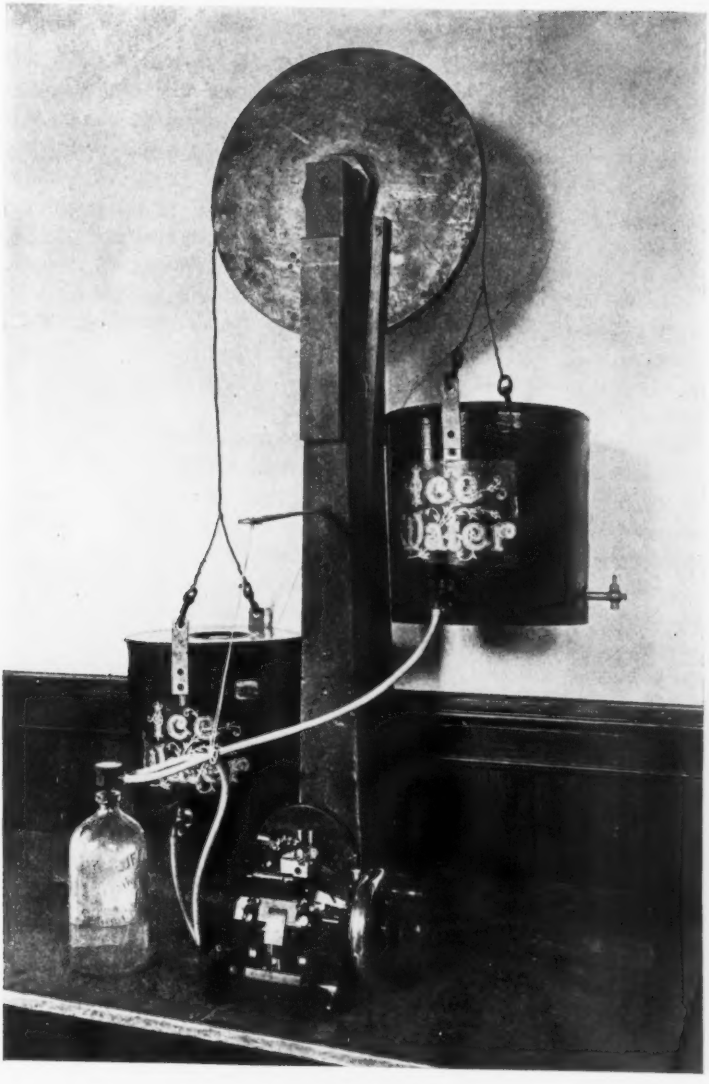

FIG. 1

the freezing mixture is put in, the 2 pails should balance. An important though simple detail is the proper adjustment of the brace to hold one of the pails up until the water runs through the tubes and freezing box into the other pail. This, as is shown in the accompanying illustration, is made of a piece of hard wood about 0.75 inch square, fastened at the upper end to the wheel by a heavy screw, the hole in the brace being large enough so that it may move freely. The brace must be long enough for its lower end to rest on the cross bar when the lower pail is about 3 inches above the base board. When the lower pail receives over half of the water it will move slowly to the base board, the lower end of the brace will pass over the cross bolt and hang perpendicularly by the side of the higher pail. As soon as this pail is emptied it should be lowered, and this should be attended to with promptness, for it is necessary that the water be kept in constant circulation to obtain the maximum freezing efficiency.

A very important part of the apparatus of course is the freezing chamber. This will have to be made to order to fit the particular microtome one is using, and the size depends upon one's needs. The one which seems to be of general use and which is employed by the writer is constructed as follows: A rod of brass about 2.25 inches long is hollowed out about 1 inch deep for the chamber, leaving walls thick enough so that a firm cap can be screwed on. The other end of the rod is trimmed down, making a stem of the desired size to fit the particular microtome. Two tubes with inside diameters 7–8 mm. and 0.75 inch long are welded into the chamber a few millimeters apart on one side. The faucets, one in each pail, should be large enough for a free flow of the water into the tubes and should be shielded on the inside of the pail by copper gauze. A second faucet should be put into one pail to be used to remove surplus water.

The freezing chamber, of course, must be in a horizontal position while the object is being frozen. A half gallon bottle with a hole in the cork large enough to receive the stem as shown in the illustration is very convenient.

It is highly desirable to have a section collector if one is cutting much material. This may be made of a block of wood about 1 inch thick, hollowed out on one side, leaving enough margin on 3 edges around the cavity so that it will fit snugly against the knife, a little vaseline being used to prevent leaking. The box may be clamped on in various ways. If this chamber is nearly filled with cold water, the sections will slide down into it and melt, after which they may all be poured out together

by removing the knife carrier and all attached thereto from the remainder of the machine. Gum arabic of medium consistency is used to freeze the objects in. A layer 2–3 mm. thick should be frozen on the chamber before placing the object on for cutting. By this means it is possible to obtain a large number of sections in a very short time, greatly facilitating the study of algae, fungi, and other soft tissues of either plants or animals in which only cell forms and cell relations are being studied. Likewise it is exceedingly useful in preparing cross and longitudinal sections of leaves, soft stems, etc., for class use. It is also inexpensive. One can run the machine 8 hours with no difficulty on 40 lbs. of ice.

In orienting the material for cutting, 2 methods may be followed. Segments of the material may be piled on top of each other on the freezing chamber and covered with gum arabic and frozen. After trimming to the desired form the material may then be removed, properly oriented, and quickly refrozen to the chamber. This is desirable only in cases in which the material is too delicate to stand on end or on edge if cross-sections are to be made. In the other method one takes the material, for example, segments of leaves a few millimeters long, dampens them in gum arabic, and piles them one upon another on a knife blade, after which the whole pile is tipped over onto smooth frozen gum on the freezing chamber. With a little care the whole pile may be made to stand on edge and may be frozen in position for cutting cross-sections.—N. L. GARDNER, *University of California.*

CURRENT LITERATURE

NOTES FOR STUDENTS

Phenomena of parasitism.—Two further contributions to a series of studies begun by BROWN[1] on the parasitism of *Botrytis cinerea* have appeared. In the first of these, BLACKMAN and WELSFORD[2] describe the microscopical details of the process of penetration of the cuticle by the germ tubes; in the second, BROWN[3] deals more specifically than in his former paper with the action on the cuticle of extracts and exudates of the germ tubes.

BLACKMAN and WELSFORD observed in the earliest stages of penetration a slight indentation of the outer epidermal wall as a result of the action of the germ tube, which is held fast to the cuticle by a mucilaginous sheath whose presence was made evident by means of a suspension of silver particles. The actual penetration of the cuticle is accomplished by a narrow peglike outgrowth from the tip of the germ tube. No swelling of the cuticle or of the subcuticular layers previous to penetration was observed, and in no case was an injury to the epidermal cells or subepidermal cells apparent before the breaking of the cuticle. Soon after the penetration of the epidermis, the cells of the palisade layer begin to disintegrate, and with the advance of the hypha the cells of the spongy parenchyma also are killed. The toxic action of the fungus extends considerably beyond the region actually invaded. After a portion of the leaf tissue had been killed, other hyphae were observed to penetrate through the stomata, probably as a result of the diffusion of food substances from the dead cells, for primary infection though a stomate was never seen.

From their observations the authors conclude that the cuticle is ruptured by mechanical pressure exerted by the germ tube and not by the solvent action of any substance secreted by it. They believe that the germ tube is enabled to exert the pressure necessary for the indentation of the cell wall and penetration of the cuticle by virtue of the gelatinuous sheath which holds the germ tube in place. It is not clear, however, how the germ tube is thus enabled to bring about an indentation of the cell wall over an area more extensive than that covered by the tip of the tube itself, as shown in some cases (notably

[1] Rev. BOT. GAZ. **61**:79. 1916.

[2] BLACKMAN, V. H., and WELSFORD, E. J., Studies in the physiology of parasitism. II. Infection by *Botrytis cinerea*. Ann. Botany **30**:389–398. *pl. 10. figs. 2.* 1916.

[3] BROWN, WM., Studies in the physiology of parasitism. III. On the relation between the infection drop and the underlying host tissue. Ann. Botany **30**:399–406. 1916.

fig. 8). It appears not improbable that these may be accidental depressions, for in many cases of actual penetration figured such indentations are not evident.

In the study of the action on the cuticle of extracts and exudates of germ tubes, BROWN found that when the extract of germ tubes was placed in considerable quantity on intact leaves and petals of *Viola*, *Petunia*, *Dahlia*, *Vicia Faba*, and *Begonia heraclaefolia*, no effect was produced; but in experiments with *Tropaeolum*, *Geranium*, *Rosa*, and *Fuchsia* a varying number of discolored spots appeared on the surfaces covered by the drops. The action in these cases was attributed to possible wounds in the cuticle. All the extracts were tested also on wounded leaves and petals, and in those cases in which no action was observed, the corresponding experiments on uninjured leaves and petals were rejected. Thus conclusions were drawn only from extracts known to be active.

When spores were sown in drops of liquid on the surface of leaves, the discoloration appeared first around the margin of the drops where the spores germinated earliest. When such drops, containing germinating spores, were displaced slightly on the leaf, the discoloration due to the action of the spores appeared within the area originally outlined by the drop and none in the new area occupied. Infection drops cleared of spores had no action on the most sensitive petals.

With reference to the possibility of the production of oxalic acid in sufficient quantity to cause the death of tissues under the uninjured cuticle, BROWN found that solutions of $n/40$ oxalic acid and of $n/20$ potassium oxalate placed on the leaves had no effect within a period of 12 hours, the time required for the germinating spores to produce discoloration. The maximum concentration in the infection drops, it was shown, could not exceed $n/800$.

These experiments seem to show quite clearly that cuticle-dissolving substances are not present in the extracts made from germ tubes of *Botrytis cinerea*, and that such substances, if they exist, do not diffuse into the surrounding medium to any considerable extent. The conclusion that chemical action is entirely excluded seems somewhat too sweeping, however, for there still remains the possibility of such action at the point of contact of the germ tube with the cuticle by substances which cannot be obtained in extracts in an active state. The possibility that oxalic acid occurs in sufficient quantities to injure cells through the cuticle seems to be definitely excluded.

The observation that the germ tubes of *Botrytis cinerea* exude no substances which are capable of diffusing through the cuticle and killing the cells below corroborates the histological study of BLACKMAN and WELSFORD, according to which the cells underlying the cuticle are not injured before the cuticle has been perforated. In this respect, the behavior of *Botrytis cinerea* differs from that of *Sclerotinia Libertiana*, in which DEBARY observed a killing of the host cells before penetration of the cuticle.—H. HASSELBRING.

Leaf size in plant geography.—RAUNKIAER,[4] whose name is associated with the system of biological types or life forms, has recently submitted another means of quantitative estimation, so far as the unit chosen is a recorder of the biological value of a climate. He regards the size of the leaf as the outstanding character, and using the simple leaf as a standard, has proposed a system of leaf classes (Bladstørrelsesklasser). In the plan submitted there are 6 different classes or divisions: (1) leptophyll, 25 sq. mm.; (2) nanophyll, $9\times25=225$ sq. mm.; (3) microphyll, $9^2\times25=2025$ sq. mm.; (4) mesophyll, $9^3\times25=18225$ sq. mm.; (5) macrophyll, $9^4\times25=164025$ sq. mm.; (6) megaphyll, which is limited by the upper limit of macrophylls. Originally he planned to use the number 10 with 25, but from a large number of trials, both by himself and several of his colleagues, 9 was found to give a better differentiation. In using 9, it is easy to make subdivisions, large, medium, and small, if desired. RAUNKIAER is of the opinion that it is an easy matter to place the various leaves in their right classes, but in order to facilitate matters, a graphical representation of the various limits of surface area is pictured, and by the use of this scheme the leaves may be correctly grouped. Thus, if a leaf has an area which is less than 25 sq. mm., it is a leptophyll; if larger than 25 sq. mm. but smaller than 225 sq. mm., it is a nanophyll, and so on.

In using such a method, RAUNKIAER contends that it is possible to obtain the biological factor for climate as far as it influences leaf size. By the use of such a scheme, comparisons may be made readily between two climates which have varying effects. One may compare formations which vary at different points, and also determine the relation between a series of associations which are somewhat similar. To prove his point he has selected and analyzed several European evergreen shrub formations.

He suggests that the leaf "size classes" are not the only quantitative units to be employed, but shows that these units lend themselves rather readily to the statistical method. A system which would in some way estimate such structural features as stomatal protection, stomatal opening, or hairiness, would also give significant results. The difficulties would naturally be many, but they should not hinder the attempt.

Ecologists and physiologists no doubt will be in hearty sympathy with RAUNKIAER's move in placing ecology upon a basis that is at least somewhat quantitative. We all are in accord with his concluding sentence (translated somewhat literally): "by such means only will it be possible to pass beyond the tourist plant geographer's superficial and vague determinations."—A. L. BAKKE.

Mountain grassland.—Many of the valleys of the Colorado Rocky Mountains have their comparatively level floors covered with grasslands of somewhat

[4] RAUNKIAER, C., Om Bladstørrelsens Anvendelse i den biologiske Plantegeografi. Botanisk Tidsskrift **34**: 225–240. 1916.

varied types, presenting ecological problems of peculiar interest. The more xerophytic type of such grassland has been studied in South Boulder Park by RAMALEY[5] and by him designated "dry grasslands" in contrast to the more mesophytic "meadow." One of the most interesting problems of the park is the relationship of these two phases of grassland, and one must regret that it has been so slightly touched upon in the present paper. Another deficiency is the limited number of data regarding the environmental factors. Some soil moisture studies seem to show that the growth water is not abundant in any association, although unfortunately the relationship of the soils of which wilting coefficient determinations were made and those whose water content were studied is not clearly apparent. Wilting coefficients ranging from 3.5 to 7.6 indicate to some extent the coarse texture and low water-retaining power of the soil which, combined with such climatic factors as short summers, high winds, and an annual rainfall of 28 inches, tend to retard the development of mesophytic vegetation.

A most interesting seasonal succession is described, ranging from a prevernal period extending from May 1 to June 15 and characterized by the blooming of *Mertensia Bakeri* and *Thlaspi purpurascens*, through well marked vernal, early and late aestival, to an autumnal in which the bloom is almost entirely limited to late grasses and blue gentian.

The series of associations involved in the xerarch succession here in progress proceeds from one characterized by *Erigeron multifidus* and *Selaginella densa* on recently exposed soil, through others in which *Carex stenophylla* associated with certain Leguminosae and Compositae such as *Aragallus Lambertii* and *Chrysopsis villosa* gradually give place to others in which grasses become increasingly abundant and important. The author regards the ultimate grassland vegetation as an association in which the grasses represented by species of *Muhlenbergia*, *Danthonia*, *Poa*, and *Festuca* predominate. Whether this will pass eventually to the more mesophytic meadow, and it in turn be replaced by forest, seems at present to be a probability not demonstrated. In spite of this and other unsolved problems, the present discussion, together with the careful analyses of the same author[6] previously published, very greatly advances our knowledge of these interesting grasslands.—GEO. D. FULLER.

Taxonomic notes.—BERRY[7] has described a new species of *Zamia* (*Z. mississippiensis*) from the Lower Eocene of Missisippi. It has "slender, graceful leaves and much reduced pinnules suggestive of *Z. floridana*."

[5] RAMALEY, F., Dry grasslands of high mountain park in northern Colorado. Plant World **19**: 249–270. *figs. 6.* 1916.

[6] ———, The relative importance of different species in a mountain grass-land. BOT. GAZ. **60**: 154–157. 1915.

———, Quadrat studies in mountain grassland. BOT. GAZ. **62**: 70–74. 1916.

[7] BERRY, E. W., A *Zamia* from the Lower Eocene. Torreya **16**: 177–179. *figs. 3.* 1916.

BLAKE,[8] in "A revision of the genus *Polygala* in Mexico, Central America, and the West Indies," recognizes 137 species, 39 of which are described as new. There are also numerous new combinations and new names, and a general reorganization of the classification.

BRITTON,[9] in connection with an account of the vegetation of "the little known island of Anegada," one of the Virgin Islands, has described a new *Acacia* (*A. anegadensis*) and a new lichen (*Arthonia anegadensis*).

BRITTON,[10] in his eighth paper on West Indian plants, describes a new *Cyperus* from Jamaica; lists the West Indian species (16) of *Stenophyllus*, including a new species; lists the Cuban species (15) of *Galactia*, with 4 new species; lists the Cuban species (5) of *Machaonia*, with 2 new species; presents the Cuban genus *Heptanthus*, recognizing 6 species, 5 of which are new; and publishes 5 new species from Porto Rico, 9 new species from Cuba, and 21 new species from the Isle of Pines, by several specialists.

BURT,[11] in continuing his studies of North American Thelephoraceae, has monographed the genus *Hypochnus*, recognizing 31 species, 13 of which are new species, and 12 are new combinations.

BURT,[12] in his seventh paper on the Thelephoraceae of North America, presents the genus *Septobasidium*. It does not belong to the Thelephoraceae, because its basidia are not simple, but it is included "merely for the convenience of students of the Thelephoraceae." The North American forms include 17 species, 10 of which are described as new.

CHRISTENSEN[13] has described a new genus (*Maxonia*) of ferns founded on *Dicksonia apiifolia* Swartz. The species (*M. apiifolia*) is represented by specimens from Jamaica and Cuba, while a variety (*M. apiifolia duale*) occurs in Guatemala, and is *Nephrodium duale* Donn. Smith.

DIXON[14] has reported upon a collection of mosses from Borneo, showing that our knowledge of the moss flora of the tropics is comparatively meager. The list includes 133 species, 13 of which are described as new. Attention is called especially to the "peculiar ecological distribution of the remarkable and striking genera *Syrrhopodon* and *Calymperes*."—J. M. C.

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[8] BLAKE, S. F., Contrib. Gray Herb. no. 47. pp. 122. *pls. 2*. 1916.

[9] BRITTON, N. L., The vegetation of Anegada. Mem. N.Y. Bot. Gard. **6**:565–580. 1916.

[10] ———, Studies of West Indian plants. VIII. Bull. Torr. Bot. Club **43**:441–469. 1916.

[11] BURT, E. A., The Thelephoraceae of North America. VI. Ann. Mo. Bot. Gard. **3**:203–241. 1916.

[12] ———, The Thelephoraceae of North America VII. Ann. Mo. Bot. Gard. **3**:319–343. *figs. 14*. 1916.

[13] CHRISTENSEN, CARL, *Maxonia*, a new genus of tropical American ferns. Smiths. Miscell. Coll. **66**:no. 9. pp. 4. 1916.

[14] DIXON, H. N., On a collection of Bornean mosses made by the Rev. C. H. BINSTEAD. Jour. Linn. Soc. Bot. **43**:291–323. *pls. 26, 27*. 1916.

Mosaic disease of tobacco.—ALLARD[15] has recently presented good evidence to combat the theory of WOODS and of HEINTZEL that oxidases are responsible for the mosaic disease of tobacco, in which he showed that the disease was dependent upon a specific infection. A more recent paper by ALLARD[16] describes in detail a study of the properties of the so-called "virus" of the mosaic disease of tobacco. Healthy plants were inoculated with the virus after filtration through a Livingston atmometer porous cup, after filtration through powdered talc, after precipitation with ethyl alcohol, after treatment with formaldehyde, with hydrogen peroxide, with precipitates of aluminum and nickel hydroxides, and after subjecting the virus to high and low temperatures. Plants were inoculated also with water extracts of the dried mosaic tobacco, made after extracting with ether, chloroform, and other solvents. The infectious principle was retained by filtration through Livingston atmometer porous cups and by powdered talc, although the filtrates gave intense peroxidase reactions. Alcoholic solutions of 75–80 per cent destroyed the infective principle, while 45–50 per cent solutions did not, but carried down the infectious principle with the precipitate. Virus treated with one part formaldehyde in 800–1500 parts of solution gave an infection. Stronger solutions gave no infection, although they still gave strong peroxidase reactions. Ether, chloroform, carbon tetrachloride, toluene, and acetone failed to extract either the infective principle or the peroxidase from dried material. The virus was killed at temperatures near 100° C., but when subjected to a temperature of −180° C. for 15 minutes it was not weakened. In every case controls were carried out with tap water and with the untreated virus. From the results the author concludes that neither enzymes nor the constituents of healthy sap can be responsible for the disease, and that since the pathogenic agent is highly infectious and capable of increasing definitely, there is every reason to believe that it is an ultramicroscopic parasite of some kind.—H. R. KRAYBILL.

Fossil Osmundaceae.—KIDSTON and GWYNNE-VAUGHAN[17] have described three species of fossil *Osmundaceae*, two of which are respectively from the Tertiary of Spitzbergen and of Queensland. Another species, *Osmundites Carnieri*, between the Tertiary and Jurassic of the Andes of Paraguay, is most interesting. The authors add something to the original descriptions of SCHUSTER from whom they received their material. The stem unfortunately is not well preserved, but the endodermis frequently joins around the margins of the leaf gaps, and an internal phloem was also probably originally present. The

[15] ALLARD, H. A., The mosaic disease of tobacco. U.S. Dept. Agric. Bull. 40. 1914.

[16] ———, Some properties of the virus of the mosaic disease of tobacco. Jour. Agric. Research **6**:649–674. 1916.

[17] KIDSTON, R., and GWYNNE-VAUGHAN, D. T., On the fossil Osmundaceae. Part V. Trans. Roy. Soc. Edinburgh **50**:469–480. *pls. 41–44.* 1916.

stem strongly resembles that of *Osmundites skidegatensis* from the western coast of Canada (Lower Cretaceous).

In a second paper, GWYNNE-VAUGHAN[18] has described the effect of injury on a narrow stele of *Osmunda regalis*. Tracheids appear in the central region of the stele. The author regards this as evidence of the stelar origin of the pith in the Osmundaceae. The voluntary blindness of British anatomists as regards medullary structures is an interesting phenomenon. They welcome the small amount of evidence which can be brought forward for the stelar origin of the pith, and close their eyes to the overwhelming evidence for its derivation from the fundamental system of tissues. The equitable procedure seems to give the same value to both kinds of evidence, and decides the question on the quantitative basis. We may record here the regret that American anatomists all feel for the untimely death of the junior author GWYNNE-VAUGHAN, whose published work is of such promise. An appreciative obituary has recently been published by SCOTT in the *Annals of Botany*.—E. C. JEFFREY.

Monomeric capsules in Bursa.—The reviewer has shown[19] that the triangular capsule of *Bursa bursa-pastoris* is produced independently by two distinct Mendelian factors (dimery), and has expressed the view (1914) that this is a derivative condition, the original form of this species probably having had only one of these factors. A considerable number of wild plants have been investigated, but as yet only one specimen has been found by the reviewer which had but one of the capsule factors, this case being still unpublished. DAHLGREN[20] has investigated a plant of this species growing in the botanical garden at Upsala, and secured from a cross with *B. Heegeri* (which lacks both of the factors for inflation of the capsules) an F_2 progeny consisting of 71 *B. bursa-pastoris* and 17 *B. Heegeri*. One of these F_2 plants produced in the F_3 16 plants having triangular capsules and 3 with turbinate capsules, thus showing that without doubt the *B. bursa-pastoris* used in this cross had monomeric capsules. There remains one important question which the author fails to mention. As *B. Heegeri* has been widely distributed in botanical gardens, it is not improbable that it had been growing in the garden at Upsala for years. If the plant used by DAHLGREN were a derivative of an earlier, *natural* cross between *B. bursa-pastoris* and *B. Heegeri*, its possession of but one of the capsule

[18] GWYNNE-VAUGHAN, D. T., On a "mixed pith" in an anomalous stem of *Osmunda regalis*. Ann. Botany **28**:351–354. *pl. 21*. 1914.

[19] SHULL, G. H., *Bursa bursa-pastoris* and *Bursa Heegeri:* biotypes and hybrids. Carnegie Inst. Washington Publ. no. 112. pp. 57. Washington. 1909.

———, Defective inheritance-ratios in *Bursa* hybrids. Verhandl. Naturf. Ver. Brünn **49**: 157–168. 1911.

———, Duplicate genes for capsule-form in *Bursa bursa-pastoris*. Zeitschr. Ind. Abstam. u. Vererbungs. **12**: 97–149. 1914.

[20] DAHLGREN, K. V. OSSIAN. Ein Kreuzungsversuch mit *Capsella Heegeri* Solms. Svensk Botanisk Tidskrift **9**:397–400. 1915.

factors would give no indication of the condition of any of the original Swedish biotypes, because plants having monomeric capsules occur normally just as frequently as those having dimeric capsules in the offspring of the F_1 and later generations from such a cross.—GEO. H. SHULL.

Liassic flora of Mexico.—WIELAND'S[21] superb quarto memoir of 165 pages and 50 plates has run the gauntlet of both the Mexican civil war and the world war, since the Spanish text has been printed in Mexico and the illustrations are from the famous lithographic firm of Werner and Winter of Frankfort. The only internal evidence of this situation is the rather large number of typographical errors in the Spanish text. The material was collected in the province of Oaxaca in the southwestern Pacific region of Mexico. For the most part it consists of impressions of leaves, and in a few instances fructifications of cycads or supposed Cycadophyta. Remains of Cordaitales are described also from the formation which is lowest Jurassic (Lias). One could wish, however, that the evidence in the case of this group were somewhat more definite, for it does not seem to establish definitely the presence of the Cordaitales in the middle Mesozoic any more than the results of LIGNIER have finally established the concurrent existence of Cordaitales and palms in the Lias of France. Only structural evidence of an unquestionable character could do this. One *Araucarioxylon* is described, but it differs from that genus in its typical form by the possession of rays of more than a single layer of cells in width. The memoir under review stands as one of the most important recent documents of systematic paleobotany in regard to the Cycadophyta, and takes its place with those of the same author on the extinct cycads of the United States and those of NATHORST on the Cycadophyta of Yorkshire, England.—E. C. JEFFREY.

Anatomy of Betulaceae.—HOAR[22] has investigated the anatomy of the Betulaceae with reference to the phylogenetic position of the family. In the Engler scheme, the family is placed among the most primitive Archichlamydeae, on the basis of flower structure. Since the most primitive family in the Engler scheme is the Casuarinaceae, the genus *Casuarina* was included in the investigation. The anatomy of the latter genus is either entirely primitive or so generalized as to include both primitive and advanced characters, so that its position near "the base of the dicotyledonous line" seems justified. The Betulaceae possess the aggregate condition of rays indicative of a primitive type, *Alnus* probably illustrating most completely the primitive condition of the family. The more advanced genera (*Carpinus*, *Ostrya*, and *Betula*) have

[21] WIELAND, G. R., La Flora Liasica de la Mixteca Alta. Bol. Inst. Geol. Mexico. no. 31. 1916.

[22] HOAR, CARL S., The anatomy and phylogenetic position of the Betulaceae. Amer. Jour. Bot. 3:415–435. *pls. 16–19.* 1916.

retained the aggregate condition only in conservative organs and regions, or it is recalled in them by injuries. The general conclusion, therefore, on the basis of anatomy, is that Betulaceae are rightly "ranked in a low phylogenetic position."—J. M. C.

Hawaiian bogs.—Situated at or near the summits of high volcanic mountains, at altitudes of 1000–2000 m., with a precipitation reaching the enormous proportions of 20 m. annually, the summit bogs of Hawaii are among the most inaccessible and remarkable in the world. In a general description of these areas MACCAUGHEY[23] calls attention to the similarity of these bogs to those of other lands in general aspect and in the presence of similar mosses, sedges, and grasses. There is an absence of many familiar forms, however, such as pitcher plants, and many of the bog ericads and orchids; while other familiar genera take new and strange forms, as instanced by woody violets and lobelias. Many endemic forms occur, particularly among the dwarf trees that form clumps scattered over the tussocky surface.—GEO. D. FULLER.

Four-lobed mother cells.—Lobed spore mother cells are very conspicuous in Jungermanniales, and by most botanists are thought to be restricted to that order. The work of ALLEN[24] adds the Musci to the list. He finds that the spore mother cells of *Catharinea* show a distinct lobing, somewhat less than in representative Jungermanniales, but nevertheless very pronounced. Lobed mother cells are present in all of the 3 orders of the Hepaticae. CAVERS reports them in *Targionia*, one of the Marchantiales; they are almost universally present in the Jungermanniales; and the reviewer finds marked lobing in the spore mother cells of species of *Anthoceros* collected by him on volcanic islets in the South Seas. The lobing of spore mother cells in bryophytes is probably of phylogenetic significance, but until much more critical work has been done it is idle to theorize.—W. J. G. LAND.

Roesleria and Pilacre.—As a result of a comparison of the various forms of *Roesleria pallida* and *Pilacre Petersii*, BAYLISS-ELLIOTT and GROVE[25] conclude, from the great similarity in structure and habit of these two fungi, that both are forms of the same plant, and that *Pilacre Petersii*, long regarded as a primitive basidiomycete of the auriculariaceous type, is therefore nothing more than the conidial form of the ascomycete *Roesleria pallida*.—H. HASSELBRING.

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[23] MACCAUGHEY, VAUGHAN, Vegetation of the Hawaiian summit bogs. Amer. Botanist **22**:45–52. 1916.

[24] ALLEN, CHARLES E., Four-lobed mother cells in *Catharinea*. Amer. Jour. Bot. **3**:456–460. *figs. 2*. 1916.

[25] BAYLISS-ELLIOTT, JESSIE S., and GROVE, W. B., *Roesleria pallida* Sacc. Ann. Botany **30**:407–414. *figs. 11*. 1916.